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Research Article

Impact of UV light exposure on the biocontrol efficacy of *Beauveria bassiana* and *Metarhizium anisopliae* against *Bactrocera zonata*

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ABSTRACT

Ultraviolet (UV) radiation and high temperatures have negative impact on persistence, pathogenicity and efficacy of entomopathogenic fungi in field conditions. This study evaluated effect of UV light, visible light and sunlight exposure on conidial viability of *Metarhizium anisopliae* and *Beauveria bassiana* at 3 intervals 15, 30 and 45 minutes. After exposure of these lights viability of conidia was evaluated through percentage germination of conidia, their pathogenicity against pupae and adult of *Bactrocera zonata.* In percentage germination data were collected at two time intervals of 24-27 hours and 48-51 hours after incubation. In pathogenicity test against *B. zonata* data of adult emergence from pupae and mortality of adult were recorded at 3, 5 and 7th day of post-treatment application. For *M. anisopliae* untreated control had showed highest conidial germination (50.83% at 24–27 hours, 87.17% at 48–51 hours) while UV-45 exposure reduced it to 16.50 % and 35.50% respectively. For *B. bassiana* germination was highest in the untreated control (30.17% and 50.83%) and lowest in UV-45 (8.50% and 22.50%). In case of *M. anisopliae* untreated control provided highest mortality of adults 62.2% at 3rd day, 80.00% at 5th day and 91.1% at 7th day. Lowest was in case of UV light treatment, 4.4% at 3rd day, 11.1% at 5th day and 13.3% at 7th day. Adult emergence was highest from pupae after UV light treatment, 57.8% at 3rd day, 68.9% at 5th day and 80.00% at 7th day. Lowest was in case of untreated control, 17.8% at 3rd day, 28.9% at 5th day and 37.8% at 7th day. In case of *B. bassiana* untreated control provided highest mortality of adults 44.4% at 3rd day, 57.8% at 5th day and 71.1% at 7th day. Lowest was in case of UV light treatment, 2.2% at 3rd day, 6.7% at 5th day and 11.1% at 7th day. Adult emergence was highest from pupae after UV light treatment, 42.2%at 3rd day, 57.8% at 5th day and 62.2% at 7th day. Lowest was in case of untreated control, 13.3% at 3rd day, 22.2% at 5th day and 26.7% at 7th day. The untreated control without light exposure gives maximum control of *B. zonata* and has a maximum pathogenicity because conidia viability was not affected with exposure to any light stress. In the comparison UV light caused more damage to the fungal conidia following sunlight and visible light respectively. Moreover, 45 minutes' exposures were most damaging than 30 and 15 minutes. This study highlighted the detrimental effect of UV light concluded that these EPF can be effective in shaded areas of orchard against *B. zonata*.

Keywords: UV light, conidial germination, Entomopathogenic fungi, Fruit fly, microbial control.

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INTRODUCTION

The peach fruit fly, *Bactrocera zonata* (Saunders) (Diptera: Tephritidae) is a major

pest of fruit and vegetables in tropical and subtropical regions of the world (Sharif et al., 2024a). Entomopathogenic fungi Beauveria *bassiana* and *Metarhizium anisopliae* have ability to cause mortality in the insect host. Several studies have reported their effectiveness against *B. zonata*. All the stages of *B. zonata* like adult and pupae were susceptible to virulent strains of *B. bassiana* and *M. anisopliae*. However, their efficacy in the field conditions is affected by abiotic components of the environment such as ultraviolet (UV) radiation. Agriculture faces important challenges such as climate change mitigation and adaptation (Hussain et al., 2024). To ensure food security and safety, authorities and farmers are focusing on sustainable agriculture and integrated pest management (Ahmed et al., 2024; Qayyum et al., 2024a). The efficiency of entomopathogenic fungus may be affected by environmental conditions, including humidity, temperature, and UV-B radiation (Quesada-Moraga et al., 2024). Also Ozone in the atmosphere entirely blocks solar radiation with a wavelength below 290nm and significantly decreases the penetration of wavelengths shorter than 320nm but nowadays due to the anthropogenic activities of humans and release of compounds like, Chlorofluorocarbons (CFCs) which are the primary cause of ozone layer damage. Solvents, spray aerosols, refrigerators, air conditioners, and other appliances commonly release them. Ultraviolet radiation breaks down chlorofluorocarbon molecules in the stratosphere, releasing chlorine atoms (Corval et al., 2021). But as now when the ozone layer depleting the ultraviolet radiation (UV) reach more on the ground which is most harmful for the existence of Entomopathogenic fungi. So, when the Entomopathogenic fungi applied on the insect pests its effectiveness, persistence, pathogenicity and shelf life decreases (Sharif et al., 2024b).

Extreme conditions with UV and high temperatures are extremely harmful to microorganisms (Qayyum et al., 2024b). The effectiveness of EPF in the field may be impacted by a number of ecological parameters, such as shortwave ultraviolet (UV) radiation from the sun, rain, temperature, humidity. Among these variables, EPF resistance to temperature extremes is thought to be especially significant since it has an impact on fungal effectiveness and persistence, as well as shelf life during storage and transportation (Shahid et al., 2024). On exposed surfaces like plant phylloplanes, sunlight especially the UV-B component is probably the most important factor in the inactivation of fungal propagules. Adding UV light protectants to conidial formulations was the first method used to counteract the effects of UV-B, however this tactic proved unproductive in many instances (Shaurub, 2023).

A technique for mitigating the harmful effects of sun radiation is to select entomopathogenic fungus strains that are more resistant to UV-B radiation. Research has shown that EPF tolerance to sun radiation varies amongst individuals (das Chagas Bernardo et al., 2020). *Metarhizium* species are more vulnerable to UV-B radiation and propagule inactivation than *Beauveria* species (das Chagas Bernardo et al., 2020). This is probably because melanin absorbs UV-B rays and functions as a sunscreen (Quesada-Moraga et al. 2024). UV-B resistance is not always increased by the presence of dark green pigments in *Metarhizium* conidia. Pigments that resemble or are made of melanin are necessary for tolerance, and *Metarhizium* species lack these pigments (Cordero et al., 2023).

In comparison to UV-A, UV-B radiation is less likely to reach the hearth surface, although being the more hazardous form of UV radiation. Ninety-five percent of solar UV light is UV-A radiation, which is softer than UV-B radiation. However, it can cause DNA damage and other indirect effects (Rai et al., 2021). Reactive oxygen species (ROS), which alter signal pathways, cause cellular toxicity, and alter genetic makeup, have been demonstrated to be produced by UV-A radiation, but in smaller amounts than by UV-B radiation (Tong and Feng 2022). The germination and survival of *M. anisopliae* conidia are adversely affected by UV-A radiation in natural conditions, emphasizing the significance of taking this into account when assessing its effects on EPF.

The visible portion of early morning light, or low-UV, is thought to help *Metarhizium* species to resist potentially fatal damage from strong UV radiation later in the day. Reactive oxygen species (ROS) are produced by UV rays in organisms, and they can cause damage to cells (Jager et al., 2017). *Metarhizium's* environmental persistence and bio insecticide efficacy could be enhanced by this method. Visible light has been shown to impact stress biology, hence more study on *Metarhizium* ecology in natural and agricultural settings is necessary (Brancini et al., 2022). Regarding *B. bassiana* and *M. anisopliae* potential as biocontrol agents against *B. zonata*, it is essential to know how UV exposure influences their effectiveness. How different intervals of UV, sunshine, and visible light exposures affect both germination and pathogenicity against *B. zonata* pupae and adults.

MATERIALS AND METHODS

Preparation of Fungal Culture

B. bassiana and *M. anisopliae* pre-maintained cultures were obtained from the Insect Pathology Lab of the Institute of Plant Protection at the MNS-University of Agriculture Multan. Colonies on freshly manufactured medium plates

were purified using a single spore purification procedure described by Zhang et al. (2013) and Noman et al. (2018). On commercially available potato dextrose agar (39 g/litre), the culture was maintained. The solution was agitated constantly at 100°C for 3-5 minutes until the homogeneous medium was completely dissolved.

The prepared media was autoclaved for 15 minutes at 121°C and 15 psi pressure to prevent contamination. The medium was autoclaved and then given some time to cool. To prevent contamination, media pouring was performed in the laminar air flow chamber, which was first UV light treated for 15 minutes and sterilized with methylated spirit for approximately 15 minutes. *B. bassiana* and *M. anisopliae* spores/conidia were inoculated onto solidified PDA plates using a sterile, autoclaved inoculation needle. To prevent contamination, the inoculated medium plates were sealed with parafilm tape strips. To acquire fungal growth, Petri Plates were cultured for 14 days in an incubator at conditions like, 25±1ºC temperature and Relative Humidity 60±10%.

Fungal Preparations

To grow the fungal culture, each fungus was inoculated on the petri plates using a metallic looping that have been heated on flame. The plates were kept in an incubator, at temperature 25±1ºC and relative humidity of 70±5% and, for 10 days. The conidia were obtained after removing from petri plates, by using a metallic spatula pre-heated on a flame. To make the suspension, sterile distilled water was added with Tween-80 at a concentration of 0.1 per ml having 1×10^8 conidia per ml.

UV and Visible Light Chambers

The specially designed chamber was made entirely of wood. The lights were positioned around 25 cm above the samples. There was an on/off switch for UV lamp and for visible light bulbs. There was a space in the chamber on the lower portion where the samples were kept. The inside temperature was tracked via a temperature sensor. A flux meter was placed in the chamber to observe the flux of the UV light. The thermometer, a door for transferring the samples, and buttons to turn on and off the lighting was all in the front section.

UV Light Exposure to Fungal Conidia

After counting the fungal spores, the effect of UV light on fungal spores was assessed by placing them in 9cm petri plates containing antibiotic at a concentration of 0.5 gL^{-1} in 100 mL of each plate for treatment, in laminar flow hood by the help of a pipette. The study included three different sources of UV light i.e. sunlight, UV lamps, visible light. The treatments include; UV lamp exposure, sunlight exposure, visible light exposure and a control that was not treated with UV light and wrapped in aluminum foil and only contain water and tween-80 with fungal concentration. Then, the Petri plates were subjected to a UV lamp radiation of around 250±5 nm range that was achieved through a 6 watt UV lamp. For visible light treatment an LED bulb of 10 watt was used. And for sunlight treatment the UV light box was placed outside under sun with the doors open where the sunlight was reached onto the treatment plates. The plates were kept away from the radiation source at 25 cm. The duration of exposure to these lights was 15, 30 and 45 minutes. A control treatment was also performed by wrapping the sample with the help of aluminum foil, to avoid the exposure of UV light. After exposure of these lights viability of conidia was evaluated through percentage germination of conidia, their pathogenicity against pupae and adult of *Bactrocera zonata.*

Conidia Percentage Germination of Conidia

Growth of fungal conidia was tested after 4, 6, and 8 days after the inoculation. To count the fungal conidia, a sample of 5 mL in each vial was used. Add 9 mL of sterile water and 1 mL of Tween-80 as a wetting agent to this sample. The suspension was quantified in hemocytometer under 400x microscope. Two 0.1 mL samples of the fungus were placed in Petri dishes with PDA culture medium to test. In percentage germination data were collected at two time intervals of 24-27 hours and 48-51 hours after incubation. Germination was recorded by examined 200 conidia at a 400x magnification, conidia was counted as germinated when the germ tube is visible.

Insect Collection and Rearing

Infested mango fruits were collected from the mango orchard in Multan, Punjab Pakistan. These infested fruits were placed on the soil in the container for the pupation of the larvae. These containers were covered with muslin cloth to avoid any other insect egg lying on the fruits. *B. zonata* larvae pupate in the container's soil. To remove the pupae from the soil, soil was sieved every three days. These pupae were shifted to a cage and placed up at the Institute of Plant Protection's Rearing Lab in the Post-Graduate Block at MNS University of Agriculture Multan. *B. zonata* culture growth was good by maintaining all rearing conditions. Light and dark period was maintained at 16:8, relative humidity was between 60±5%, and temperature was 27±2 °C. Adults of fruit fly was reared onto the sugar and water solutions that was added onto the absorbent cotton in small cups.

Pupal Bioassay

Bioassay on pupae of *B. zonata* was carried out to access the pathogenicity of fungal conidia. For the bioassay 3 day old pupae were used. Using the immersion method, conidia was assessed against a batch of 15 *B. zonata* pupae. Pupae were dipped for 15 sec in Petri dish with a concentration of 1×10^8 spore's mL^{-1}. The treatments used against pupae were; UV light, visible light, sunlight and a control of untreated control against both *B. bassiana* and *M. anisopliae* with 3 time durations each 15, 30, 45 (UV-15, UV-30, UV-45, VL-15, VL-30, VL-45, SL-15, SL-30, SL-45, untreated control). So, the total treatments were 10 with 3 replications of each treatment by using the complete randomized design (CRD). The adult emergence was evaluated at 3, 5, and 7 days after treatment. And the bioassay stopped after 8 days from the start of bioassay. For treatments light and dark period was maintained at 16:8, relative humidity was between 60±5%, and temperature was 27±2 °C.

Adult Bioassay

After evaluation of conidial viability, bioassay on adult of *B. zonata* was carried out to access the pathogenicity of fungal conidia. For bioassay of adult small sterilized boxes of 23 cm height and 9 cm diameter were used. 1 day old adult of *B. zonata* were treated with *B. bassiana* and *M. anisopliae.* Conidial pathogenicity was assessed against a batch of 15 *B. zonata* adults. For adult a concentration of 1×10^8 conidia mL^{-1} was used, 1ml of the same concentration was sprayed on to the adult colony using hand atomizers. The treatments used against adult were; UV light, visible light, sunlight and untreated control against both *B. bassiana* and *M. anisopliae* with 3 time durations each 15, 30, 45 (UV-15, UV-30, UV-45, VL-15, VL-30, VL-45, SL-15, SL-30, SL-45, Untreated control). So, the total treatments were 10 with 3 replications of each treatment by using CRD. The adults were fed on artificial diets when in bioassay boxes. The diet used was sugar solution and water solution in small diet cups by soaking absorbent cotton. The mortality of the adult was evaluated at 3, 5, and 7 days after treatment. And the bioassay stopped after 8 days from the start of bioassay. For treatments light and dark period was maintained at 16:8, relative humidity was between 60±5%, and temperature was 27±2 °C.

Statistical Analysis

Abbott formula was used to calculate the corrected mortality (Abbott, 1925). The collected data was analyzed using STATIX 8.1 to determine significance of all treatments means. The analysis was carried out using Analysis of Variance under completely randomized design to compare the means at the 5% probability level. Tukey's HSD was used in multiple comparison tests to differentiate in means in different treatments.

RESULTS

Percentage Germination of Conidia

M. anisopliae (*Ma*) conidia germination varied significantly depending on the light exposure. At the first observation interval (24-27 hours), the germination of *Ma* conidia differed greatly depending on the light exposure conditions. The untreated control has the highest germination rate, around 55%, demonstrating best conditions for conidial viability in the absence of light stress. Among the treated groups, visible light (VL) exposure resulted in greater germination than sunlight (SL) and ultraviolet (UV) treatments. Particularly, Ma-VL15 had the greatest germination rate out of all VL treatments, attaining nearly 50%, followed by Ma-VL30 (40%) and Ma-VL45 (35%). Sunlight exposure had a moderate influence on germination, with Ma-SL15 achieving 32%, Ma-SL30 at 30%, and Ma-SL45 at 25%. whereas, UV radiation lowered germination greatly, with Ma-UV15 at 27%, Ma-UV30 at 22%, and Ma-UV45 at just 15%. These findings showed that UV light had an immediate negative impact on conidial germination, whereas VL exposure supported significantly higher germination within the first 24-27 hours.

At the next observation interval (48-51 hours), all treatments showed improvements in germination; however the degree of change differed based on the kind of light exposure. The untreated control had the highest germination rate, reaching almost 90%, proving the natural germination capacity of Ma conidia in the absence of light stress. Among the VL treatments, Ma-VL15 had the greatest germination rate at about 85%, followed by Ma-VL30 at 80% and Ma-VL45 at 70%, indicating that VL exposure had a comparably good effect on conidial viability over time. Sunlight-exposed conidia also shown increased germination, with Ma-SL15 arising to 65%, Ma-SL30 to 60%, and Ma-SL45 to 55%, yet these results were below compared to those obtained from VL treatments. UV exposure maintained to have the inhibitory effects, with Ma-UV15 emerging to 50%, Ma-UV30 to 43%, and Ma-UV45 to 35%. Conidial germination percentage (%) was showed in the sequence of: Untreated Control > Ma-VL15 > Ma-VL30 > Ma-VL45 > Ma-SL15 > Ma-SL30 > Ma-SL45 > Ma-UV15 > Ma-UV30 > Ma-UV45 (Figure. 1).

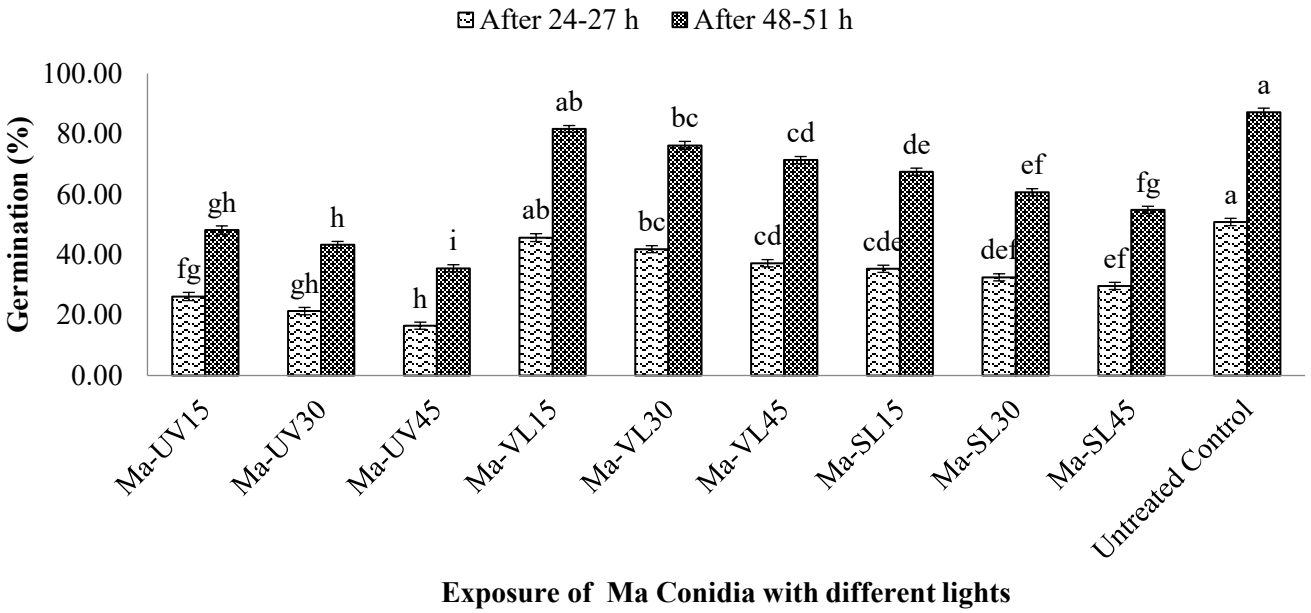

Figure 1. Germination (%) of M. anisopliae conidia exposed to UV, visible light (VL), and sunlight (SL) for 15, 30, and 45 min, assessed at 24–27 h and 48–51 h post-incubation
Bars with the same letter are not significantly different (Tukey's HSD, $p < 0.05$)

Same trend was observed for conidial germination percentage (%) of *B. bassiana*: Untreated control > Bb-VL15 > Bb-VL30 > Bb-VL45 > Bb-SL15 > Bb-SL30 > Bb-SL45 > Bb-UV15 > Bb-UV30 > Bb-UV45. In case of *B. bassiana* the highest conidial germination percentage (%) was 30.17% after 24-27 hours and 50.83% after 48-51 hour for untreated control of *B. bassiana*. And lowest was in Bb-UV45, 8.50% after 24-27 hour and 22.50% after 48-51 hour. The results show that conidial germination percentage (%) for *M. anisopliae* was more than *B. bassiana* (Figure. 2). Although an improvement in germination over time, UV-exposed conidia had much lower viability than the other treatments. These data indicate that, whereas conidia may recover from light-induced stress, continuous UV exposure significantly inhibits their germination capability, whereas VL exposure maintains conidial viability with time.

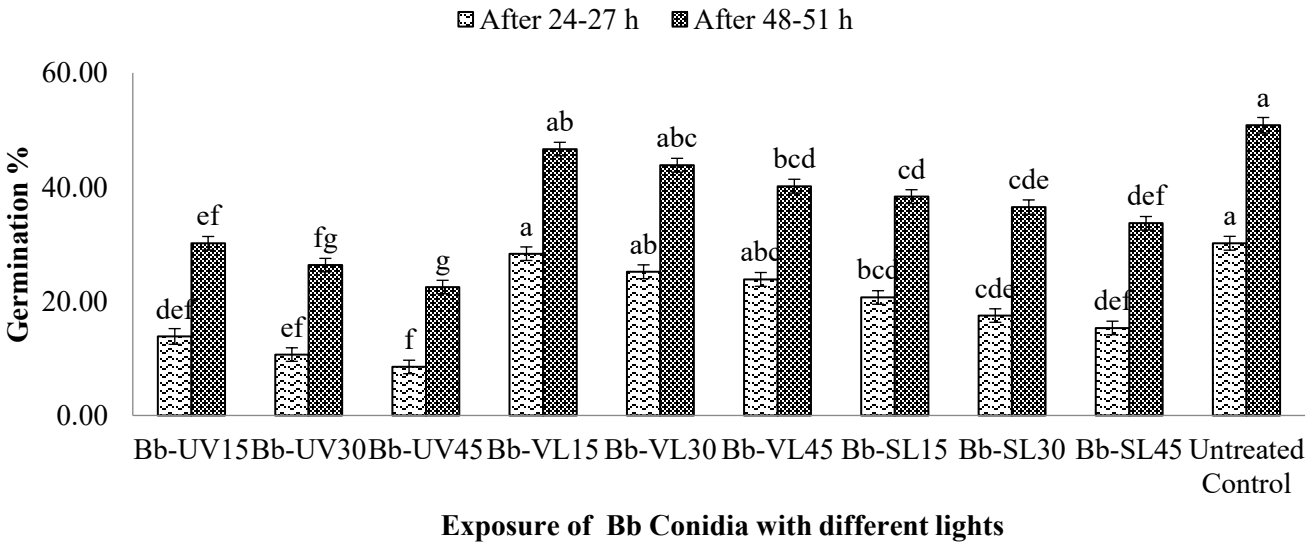

Figure 2. Germination (%) of *B. bassiana* conidia exposed to UV, VL, and SL for 15, 30, and 45 min, assessed at 24–27 h and 48–51 h post-incubation
Bars with the same letter are not significantly different (Tukey's HSD, $p < 0.05$)

Adult Bioassay

The adult mortality of *B. zonata* was recorded at 3, 5 and 7th day of and *M. anisopliae* conidia application. The untreated control group showed the highest level of mortality, achieving about 60% at 3 days, 75% at 5 days, and 85% at 7 days, considerably more than all Ma-treated groups ($P < 0.05$). Of all the treatments, the greatest mortality was recorded in visible light exposure. In the Ma-VL15 treatment, mortality was about 50% at 3 days, 65% at 5 days, and 75% at 7 days, while Ma-VL30 showed similar findings with mortality about 52%, 67%, and 76% at the each intervals. Ma-VL45 showed a decreased mortality nearly 45%, 60%, and 70% over the three-time intervals.

In comparison, UV-exposed groups had considerably reduced mortality rates. Ma-UV15 caused about 15%, 20%, and 25% mortality at 3, 5, and 7 days accordingly, whereas Ma-UV30 showed mortality rates of around 10%, 18%, and 22% during the same period. The Ma-UV45 group had the lowest mortality rate, with values of around 5%, 12%, and 18% at the various time periods. Similarly, sunshine exposure reduced the effectiveness of Ma conidia. Ma-SL15 caused 35% mortality at 3 days, which increased to 50% at 5 days and 65% at 7 days. Ma-SL30 had somewhat lower mortality rates of 20%, 30%, and 45% at 3, 5, and 7 days, respectively. The lowest mortality rates in sunlight-exposed group recorded in Ma-SL45, with 10%, 15%, and 22% respectively. Adult mortality percentage was in the sequence of: Untreated control > Ma-VL15 > Ma-VL30 > Ma-VL45 > Ma-SL15 > Ma-SL30 > Ma-SL45 > Ma-UV15 > Ma-UV30 > Ma-UV45 (Figure. 3). The results suggested that visible light maintained the pathogenicity of conidia, but UV and prolonged sunlight exposure considerably decreases its effectiveness. These findings showed the need of regulating conditions for the efficient use of entomopathogenic fungi in biological management of *B. zonata*.

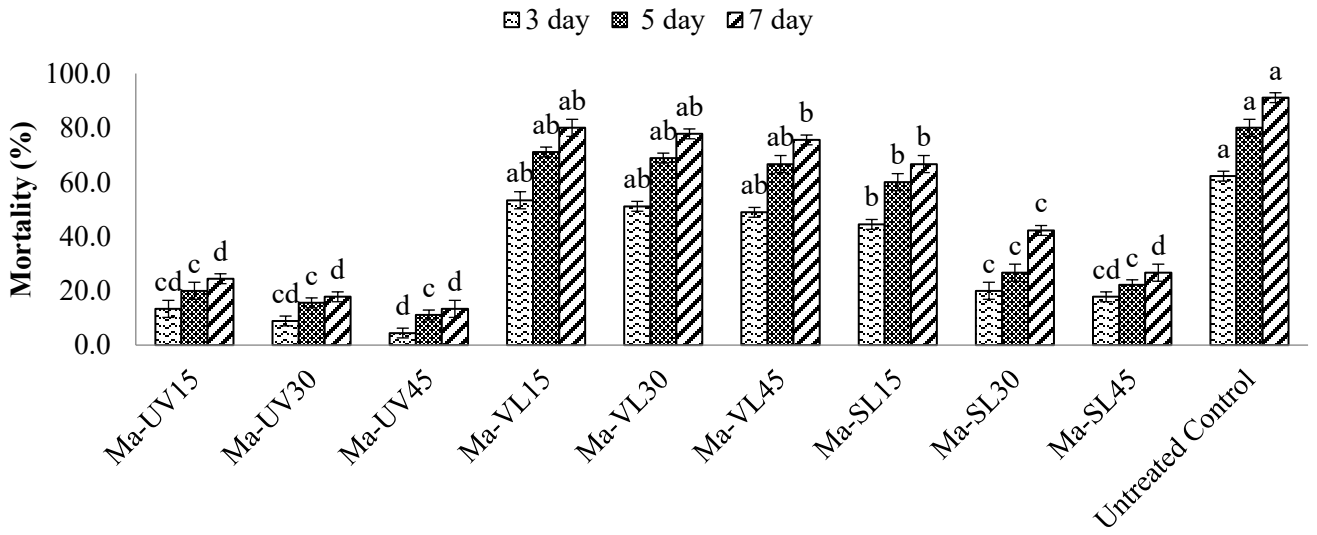

Figure 3. Adult mortality (%) of *B.* zonata treated with *M. anisopliae* conidia exposed to UV, VL, and SL for 15, 30, and 45 min. Mortality recorded at 3, 5, and 7 days post-treatment
Different letters indicate significant differences (Tukey's HSD, $p < 0.05$)

Similar trend in the adult mortality of B. zonata was observed in the case of *B. bassiana* as: Untreated control > Bb-VL15 > Bb-VL30 > Bb-VL45 > Bb-SL15 > Bb-SL30 > Bb-SL45 > Bb-UV15 > Bb-UV30 > Bb-UV45. In case of *B. bassiana* the highest % adult mortality was 44.4% after 3 days, 57.8% after 5 days and 71.1% after 7 days for untreated control. Lowest adult mortality percentage was recorded in Bb-UV45 as 2.2% after 3 days, 6.7% after 5 days and 11.1% after 7 days (Figure. 4).

Pupal Bioassay

At 3, 5, and 7 days adult emergence percentages for Ma-UV15 were approximately 50%, 60%, and 72%, respectively, while Ma-UV30 had 52%, 63%, and 75% emergence. Ma-UV45 had the highest emergence rate among UV-exposed groups, with values reaching 58%, 72%, and 80%, indicating that increased UV exposure was the cause of the higher emergence rates. Compared to UV-treated groups, pupae exposed to Ma conidia under visible light had considerably reduced emergence rates. At 3, 5, and 7 days, emergence rates were 30%, 40%, and 52% for Ma-VL15, 35%, 45%, and 58% for Ma-VL30, and 40%, 50%, and 65% for Ma-VL45.

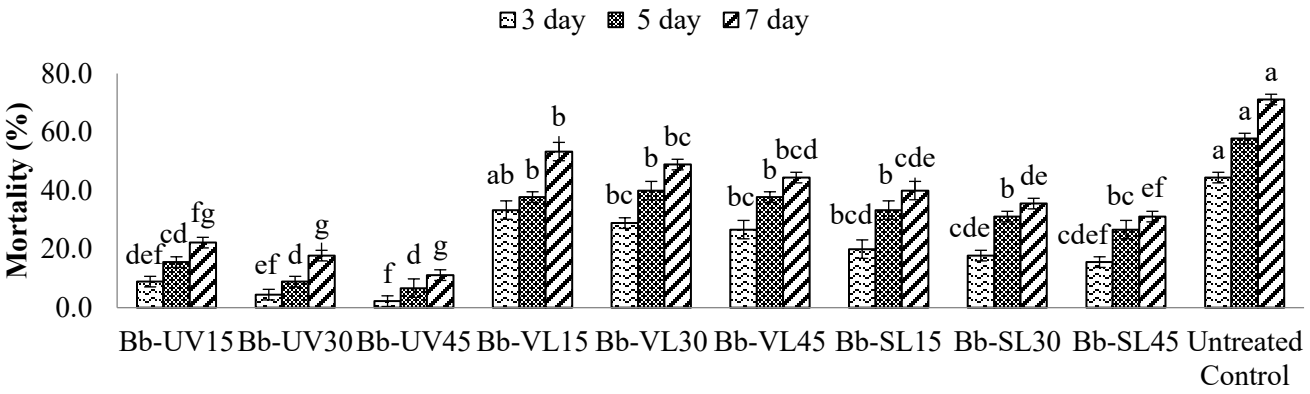

Figure 4**.** Adult mortality (%) of *B. zonata* treated with *B. bassiana* conidia exposed to UV, VL, and SL for 15, 30, and 45 min. Mortality recorded at 3, 5, and 7 days post-treatment
Different letters indicate significant differences (Tukey's HSD, $p < 0.05$)

These findings suggest that visible light exposure preserves Ma conidia effectiveness better than UV exposure. The Ma-SL15 treatment demonstrated emergence rates of 38%, 48%, and 60% at 3, 5, and 7 days, accordingly, while Ma-SL30 showed 42%, 52%, and 65%. The Ma-SL45 group reported emergence rates of 45%, 55%, and 68%, showing that extended sunlight exposure led to moderate loss of EPF effectiveness. The untreated control group demonstrated the least adult emergence, with 10%, 18%, and 30% at 3, 5, and 7 days, accordingly These results demonstrate the major pathogenic effect of Ma conidia against *B. zonata* and indicate the adverse effect of UV and sunlight exposure on EPF effectiveness. Adult emergence percentage was in the sequence of: Untreated control < Ma-VL15 < Ma-VL30 < Ma-VL45 < Ma-SL15 < Ma-SL30 < Ma-SL45 < Ma-UV15 < Ma-UV30 < Ma-UV45 (Figure. 5). Similar trend of adult emergence percentage was observed when pupae were treated with conidia of *B. bassiana*: Untreated control < Bb-VL15 < Bb-VL30 < Bb-VL45 < Bb-SL15 < Bb-SL30 < Bb-SL45 < Bb-UV15 < Bb-UV30 < Bb-UV45. In case of *B. bassiana* the lowest adult emergence percentage was 13.3% after 3 days, 22.2% after 5 days and 26.7% after 7 days for untreated control. Highest adult emergence percentage was in Bb-UV45, 42.2% after 3 days, 57.8% after 5 days and 62.2% after 7 days (Figure. 6). Those results demonstrate that EPF conidia were more efficient under visible light conditions and extensively degraded under UV and sunlight exposure, highlighting the need for protective formulations or application strategies that reduce the influence of light exposure to enhance the effectiveness of EPF in the biological control of *B. zonata*.

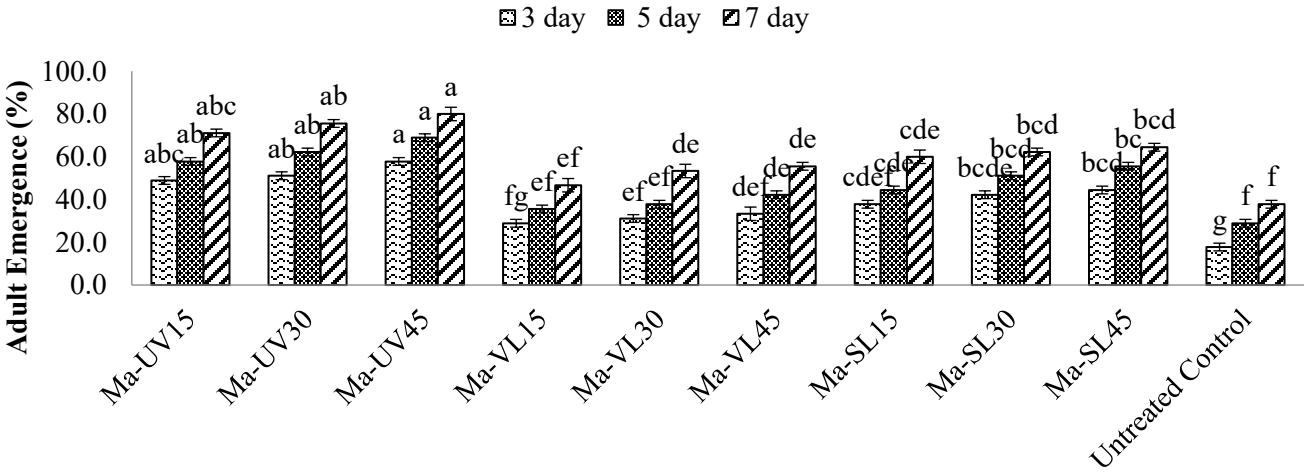

Figure 5. Adult emergence (%) from B. zonata pupae treated with M. anisopliae conidia exposed to UV, VL, and SL for 15, 30, and 45 min. Emergence assessed at 3, 5, and 7 days post-treatment
Different letters indicate significant differences (Tukey's HSD, $p < 0.05$)

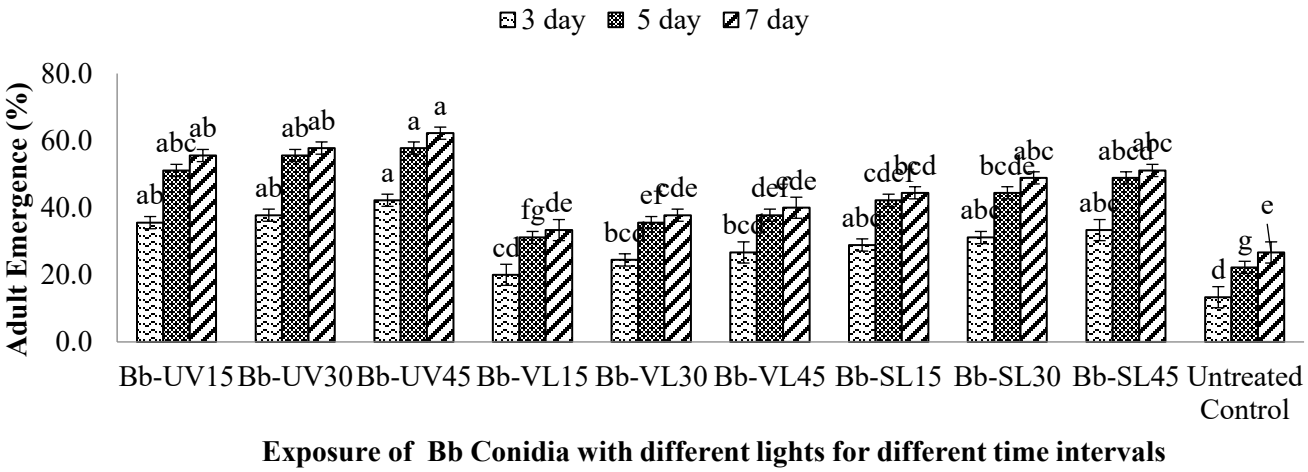

Figure 6. Adult emergence (%) from *B. zonata* pupae treated with *B. bassiana* conidia exposed to UV, VL, and SL for 15, 30, and 45 min. Emergence assessed at 3, 5, and 7 days post-treatment
Different letters indicate significant differences (Tukey's HSD, $p < 0.05$)

DISCUSSION

In this study the conidia of two Entomopathogenic fungi *M. anisopliae* and *B. bassiana* were exposed with UV light, Visible light and sunlight to check the viability of conidia and germination percentages. The conidia were exposed, and viability checked after 24-27 hour and 48-51 hour post-exposure. UV light exposed conidia caused loss in germination percentage in both *M. anisopliae* and *B. bassiana*. These results are same in comparison to previous studies conducted which results in delayed germination at 24–48-hour post-exposure to UV light of both EPF's *M. anisopliae* and *B. bassiana* (Braga et al., 2001d; Posadas et al., 2012; Birnbaum et al., 2021).

UV light exposure significantly reduces germination and growth, particularly with longer exposure times as was in this study at 45 minutes exposure this was in comparison to previous studies (Costa et al., 2012; Biryol et al., 2021). According to a study Exposure to UV light or sunlight quickly inactivated *B. bassiana* conidia (Edgington et al., 2000). For every formulation, the mean germination level decreased with an increase in the length of exposure to simulated solar radiation. It is evident that UV exposure delays germination because control plates of all formulations that were not exposed to simulated sunshine showed higher mean conidial germination than plates subjected to simulated sunshine. Although the cause of the surviving conidia's delayed germination is unknown, it could be directly related to damage to proteins and nucleic acids, which would slow down growth, or it could be a result of the conidia's defensive responses, or may reflect the time spent or energy diverted to repair damage (das Chagas Bernardo et al., 2020). These results are in conformity to our research where untreated control as a control gives highest percentage of germination almost 87.17% in case of *M. anisopliae* and 50.83% in case of *B. bassiana* after 48-51 hours of exposure. Sunlight and UV rays negatively impact the viability of entomopathogenic fungi, reducing infection rates. Radiation negatively impacts entomopathogenic fungal performance (Fang et al., 2012). This is in comparison to our study in which sunlight and UV light limit the effectiveness of both Entomopathogenic fungi.

Another study revealed that main ways that UV light harms entomopathogenic fungi are by oxidative denaturation of proteins, membrane peroxidation, and direct DNA damage (resulting in the synthesis of cyclobutane pyrimidine dimers and 6-4 photoproducts) (Tong and Feng, 2022). Furthermore, *B. bassiana* is more susceptible to UV-B rays than *M. anisopliae* due to its lack of melanin pigmentation. This demonstrates that *M. anisopliae* showed more tolerance in our research because to its dark green colored conidia and photo-reactivation restoration mechanisms (Acheampong et al., 2020). The results are in confirmation to the previous studies which states that UV resistance resulted from non-enzymatic and enzymatic defense mechanisms. Enzymes superoxide dismutase and catalase reduce oxidative damage caused by UV light and heat. Pigment accumulation provides non-enzymatic defense and protects against oxidative stress (Rangel et al., 2006). The dark green conidial pigmentation of the *M. anisopliae* isolates should have provided more UV protection by inhibiting radiation in compared to the hyaline, light-pigmented *B. bassiana* conidia (Braga et al., 2006; Fang et al., 2010; Costa et al., 2021; Corval et al., 2021). Photo reactivation is a significant repair process found in the *Metarhizium* genus that helps to repair DNA damage caused by UV radiation (Fang and Leger, 2012).

Our findings were aligned with the previous studies in which *B. bassiana* is more susceptible to UV-B rays than *M. anisopliae* (Braga et al., 2001; Falvo et al., 2016; Shahid et al., 2024). Rapid inactivation of fungal conidia of *B. bassiana* due to effect of UV-B has been reported (Edgington et al., 2000; Hemalatha et al., 2017). This research can be beneficial for the farmer community regarding developing application strategy of biopesticide, avoiding exposure of sunlight and for the biopesticide formulation industry to improve formulation to reduce the damaging effects of these radiations. This study was more focused on one abiotic component that was exposure of light. In future studies effects of humidity level and temperature can also be studied. In future there is need to explore the overexpression of DNA repairing and melanin production mechanisms, identification of biomarkers for stress tolerance in entomopathogenic fungi and development of the stress tolerant formulations.

CONCLUSIONS

UV light exposure significantly reduces germination and growth of entomopathogenic fungi (EPF) that affects its biocontrol efficacy. So, apply EPF early morning, evening and in shaded areas to avoid exposure of these lights. UV protectors in the formulations should be used to minimize UV induced inactivation of EPF. Combine EPF with other IPM strategies like sanitation and traps for management of *B. zonata*.

AUTHOR CONTRIBUTIONS

Mirza Abdul Qayyum concealed the idea and supervised the research. Muhammad Zeeshan Shahid conducted the research trial and collected data. Saira Akhtar performed statistical analysis. Umer Sharif help in research trials and wrote this manuscript. Hasan Taha, Khuram Shahzad and Muhammad Aali Shan help in data collection and examination of the trials regularly. Mirza Abdul Qayyum and Muhammad Ishtiaq reviewed the manuscript.

COMPETING OF INTEREST

No conflicts of interest have been disclosed by the authors.

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