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Research Article

Effect of Ultraviolet (UV) Light on Cowpea Beetle *Callosobruchus maculatus* Fabricius (Coleoptera: Chrysomelidae) under Laboratory Conditions

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AAP conducted the experiment and wrote initial draft. AAG designed the study. LBR and JGMS designed and supervised the study. MIM and AAG finalized the manuscript. MIM revised the manuscript. FNG analysed and presented the data.

Keywords
Cowpeas, Light, Management, Pulse beetle, Stored grains, Ultraviolet

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Abstract | *Callosobruchus maculatus* is a cosmopolitan and severe pest of pulses throughout the world with significant damage potential. Mostly synthetic fumigants are used to reduce population and losses of *C. maculatus,* but their use causes serious health and environmental problems, hence necessitating the use of alternate safe control measures for its management, including ultraviolet light radiation (UVL). Therefore, laboratory studies were conducted to determine the effect of different powers i.e., 15, 30, and 45 watts of UVL against eggs and adults of *C. maculatus* applied for 1, 2, 3, 4, and 5-h exposure times. UVL tube-lights of 15 watt power and 1.5 feet long were installed in 2-feet^3 insect rearing cages, covered with black cloth. The experiment was arranged in a completely randomized design with five replications. The data on adult mortality of *C. maculatus* was taken after 24, 48, and 72-h of treatment, whereas hatching of eggs was monitored daily for ten days. Results indicate a highly significant difference among various UVL powers and exposure timings on the mortality of *C. maculatus* adults as the mortality percentage increases with increasing exposure timings and powers with no significant difference between 30- and 45-watt UVL powers. After 72-h of the experiment, the highest adult mortality (74.80±2.80%) was observed with 45-watt UVL, followed by 30-watt UVL (65.20±1.92%), both applied for 5-h. Moreover, the highest adult mortality rate of *C. maculatus* adults was recorded when they were exposed to various UVL power for 5-h. Like adult mortality, the lowest hatching success of *C. maculatus* eggs was recorded in 45-watt UVL power (27.40±2.71%), whereas the maximum eggs hatched in control treatment (88.00±0.90%), whereas 5-h exposure of *C. maculatus* eggs with different UVL powers resulted in the minimum hatching success. Therefore, it is recommended that UVL irradiation should be included in the management of *C. maculatus* in warehouses, whereas further studies should be conducted to optimize appropriate power and exposure timings of UVL for the large-scale use on commercial basis.

Novelty Statement | Laboratory evaluations of UVL different exposures and timings against *Callosobruchus maculatus* confirmed that UVL can cause significant mortality of adults and reduce the hatchability of the eggs. As a result, UVL resulted in low population of *C. maculatus* that resulted in low damage to cowpea grains without using synthetic insecticides.

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Introduction

The cowpea beetle, *Callosobruchus maculatus* (F.) (Coleoptera: Chrysomelidae) is a global and severe pest of stored pulses (Naseri and Hamzavi, 2021). *Callosobruchus maculatus* is a very destructive and damaging insect pest of commonly consumed pulses such as chickpea, cowpea, and lentils as its feeding can result up to 67% losses of the pulses (Jaiswal *et al*., 2019; Babu *et al*., 2021). Generally, its larvae cause significant losses feeding on seed embryo of the pulses, whereas adults typically do not feed (Keita *et al*., 2001; Bidar *et al*., 2021; Mushawwir *et al*., 2023). Among various management strategies, synthetic pesticides remain the most widely used and dependable approach for controlling *C. maculatus*, primarily due to their high efficacy, low cost, and ease of application (de Andrade Rodrigues *et al*., 2022). In warehouse storage systems, pyrethroids, organophosphates, and phosphine-based insecticides are predominantly employed to suppress *C. maculatus* populations (Iturralde-Garcia *et al*., 2016). However, the extensive and indiscriminate use of these chemical agents has resulted in serious concerns related to human health and environmental safety (Wakil *et al*., 2013; Pourya *et al*., 2018), as well as the development of resistance in *C. maculatus* populations to commonly used insecticides (Malaikozhundan and Vinodhini, 2018; Mutlu *et al*., 2019). Consequently, there is an urgent need to explore alternative control strategies that ensure sustainable management of *C. maculatus* in stored pulses while maintaining safety for humans and the environment (de Andrade Rodrigues *et al*., 2022).

Among the alternative measures, ultraviolet light radiations have been evaluated against stored grain pests including *C. maculatus* (Kalpna *et al*., 2022; Sahadia and Aziz, 2011). Ultraviolet radiations (UV) have been reported as germicidal in the insect management (Arul *et al*., 2022), as it is proved disinfectant towards insect eggs when applied at surfaces, whereas an attractant for adult beetles, affecting their physiological, embryological, and gene expression aspects (Arul *et al*., 2022; Bhardwaj *et al*., 2019; Dudi *et al*., 2023). Accordingly, UV radiations have also been used against various stored-grain pests such as *Sitophilus zeamais* (Tungjitwitayakul *et al*., 2016; Ferreira *et al*., 2018), *Trogoderma granarium* (Masoumi *et al*., 2021), *S. oryzae* and *Tribolium castaneum* (Draz *et al*., 2021), *Rhyzopertha dominica* (Srivastava *et al*., 2019) and others. All the studies recorded significant impact of UV radiation on mortality and reproductive parameters of these pests as mainly their effectiveness increased with higher exposure timings (Kalpna *et al*., 2022). Moreover, the UV light has also been found effective against *Callosobruchus* spp., affecting its various biological parameters (Bhardwaj *et al*., 2019; Heidari *et al*., 2016).

Therefore, considering the effectiveness of UV light against *Callosobruchus* spp., studies were conducted to determine the impact of UV lights applied at various exposure timings against *C. maculatus* under laboratory conditions, as the results obtained can be helpful to manage its loss with less use of synthetic insecticides in pulses.

Materials and Methods

Experiments were conducted in the Stored Grain Research Laboratory, Department of Entomology, Sindh Agriculture University, Tandojam, Sindh, Pakistan. The initial culture of *C. maculatus* was obtained from the culture reared in the laboratory from collected samples from Mirpur Khas population during the survey study. The F_1 population obtained from this culture was used in the experiment.

The source of UVL irradiation used in the study were ultraviolet tube lights, Sogo F-15T8/GL (Beijing, China), each having a power of 15watts, measuring 1.5 feet long, and emitting a wavelength of 254 nm (UV). The tube lights were installed at the ceiling of a 2*2*2 feet insect rearing cage. The targeted *C. maculatus* adults and eggs, kept in petri dishes were placed at the bottom of the cages (Figure 1).

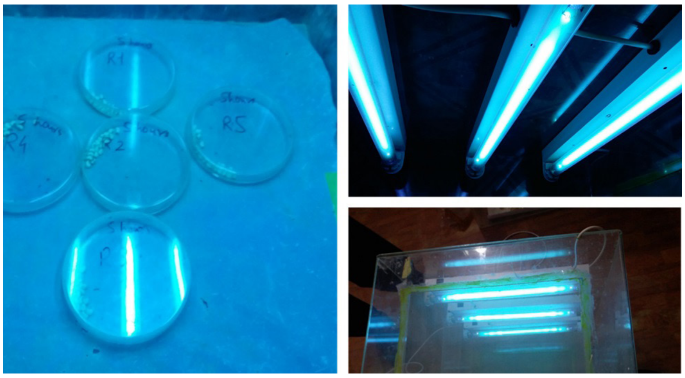

Figure 1: Experimental set-up of UV lights against *Callosobruchus maculatus* adults and eggs.

Five pairs of five newly emerged pairs (24 h old) of *C. maculatus* were kept in petri dishes with cowpea grains to assess the effect of UV light, while twenty freshly laid eggs, each on a single seed per cowpea grain, were used to record hatchability. Three UV light intensities, 15, 30, and 45 watts, were applied by turning on one, two, and three tube lights, respectively. Both adults and eggs were exposed to different UV light powers for varying durations: 1, 2, 3, 4, and 5 h. After each exposure, the adults and eggs were immediately removed from the cage and kept in the laboratory under standard conditions (30 ± 2°C and 60 ± 5% RH). Control groups for both adults and eggs were also maintained in the same cage with similar exposure times to evaluate the effects of the cage environment on mortality and egg hatching.

Mortality of adults was recorded after 24, 48, and 72-h of exposure, whereas hatching of eggs was monitored daily for ten days to ensure that the maximum number of eggs had been hatched in the control treatments. Following equations were used for the calculation of adult mortality and hatching of eggs:

$$\text{Adult Mortality \%} = \frac{\text{Dead adults}}{\text{Adults irradiated}} \times 100$$

$$\text{Egg hatching \%} = \frac{\text{Eggs hatched}}{\text{total eggs irradiated}} \times 100$$

The experiment was arranged in a Completely Randomized Design (CRD) where each treatment combination was replicated five times. ANOVA was used for the analysis, whereas means with significant differences were separated using the LSD. The STATISTIX 8.1 computer software was used for the analysis.

Results

The effect of Ultraviolet (UV) light on C. maculatus *adults*

Figure 2 illustrates results regarding the impact of various UV light powers applied at various exposure timings against *C. maculatus* after 24-h of application. A highly significant difference was recorded among various UV light powers applied at 1, 2, 3, 4, and 5 h (F = 3.98, P < 0.001) to cause mortality among targeted *C. maculatus* adults as a gradual rise in the mortality of adults was recorded with increased exposure timings. Significantly the highest percentage mortality of adults i.e., 50.00±6.30 and 42.00±5.80% was recorded at 45- and 30-watt power, respectively at the exposure time of 5-h. The highest mortality percentage of adults recorded at 15-watt UV light was 24.00±2.40% when exposed for 5-h. No mortality was recorded in the control treatment at all the exposure timings.

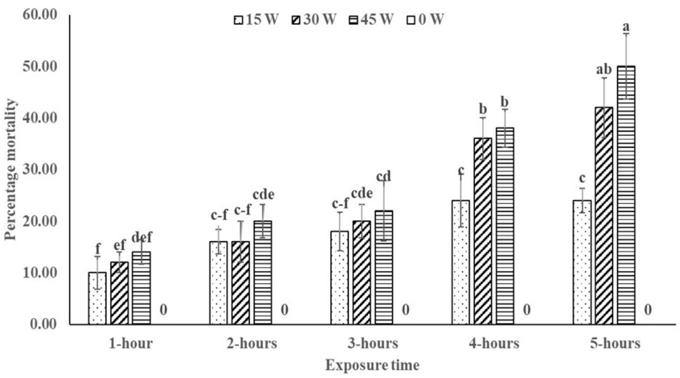

Figure 2: Effect of ultraviolet light radiation powers at various exposure intervals on mortality percentage of *Callosobruchus maculatus* adults after 24-h.
***Means followed by same letters are not significantly different from each other (LSD = 9.870, P < 0.05).**

The mortality data recorded after 48-h of the application of various exposure timings of UV light against *C. maculatus* adults showed a further rise in their mortality rate, however, no significant difference was observed in UV-light watts applied for various exposure timings (F = 1.01, P = 0.04435) (Figure 3). according to the results, the maximum mortality percentage recorded at 15-, 30-, and 45- watts recorded was 54.00±5.10, 66.00±4.00, and 68.00±5.80%, respectively, all observed when UV lights were applied for 5-h. Moreover, the minimum mortality at all these UV light powers was 36.00±4.00, 42.00±3.70, and 48.00±3.70%, respectively, all observed at 1-hour exposure timing.

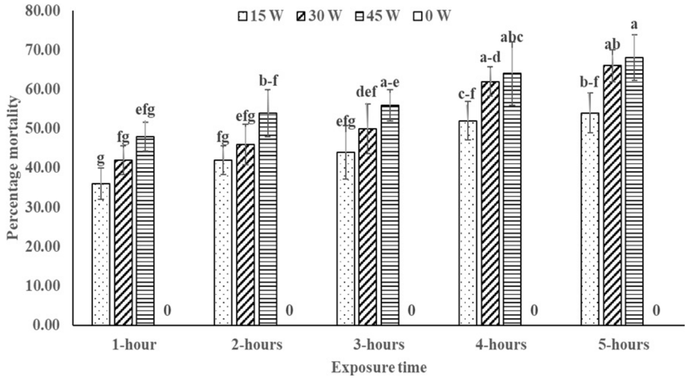

Figure 3: Effect of ultraviolet light radiation powers at various exposure intervals on mortality percentage of *Callosobruchus maculatus* adults after 48-h.
***Means followed by same letters are not significantly different from each other (LSD = 12.618, P < 0.05).**

Like 48-h data, after 72-h of observation, no significant difference (F = 1.02, P = 0.4414) was observed in the effectiveness of various UV light powers applied at five exposure timings to elicit the mortality among targeted *C. maculatus* adults. Accordingly, the highest adult mortality (86.00±6.00%) was recorded at 45-watt power UV light applied for 5-h that was not significantly different from percentage mortality of 80.00±4.50% recorded in the same power UV light applied for 4-h. Moreover, maximum percentage adult mortality recorded in 30- and 15-watt UV light was 74.00±2.40 and 68.00±7.30%, respectively, both observed at 5-h exposure timing (Figure 4).

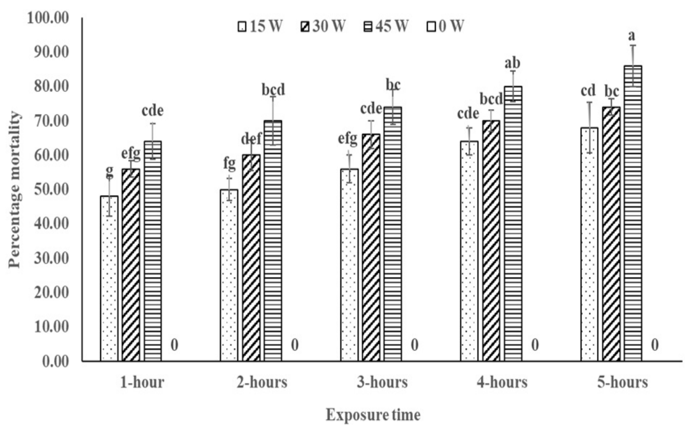

Figure 4: Effect of ultraviolet light radiation powers at various exposure intervals on mortality percentage of *Callosobruchus maculatus* adults after 72-h.
***Means followed by same letters are not significantly different from each other (LSD = 11.706, P < 0.05).**

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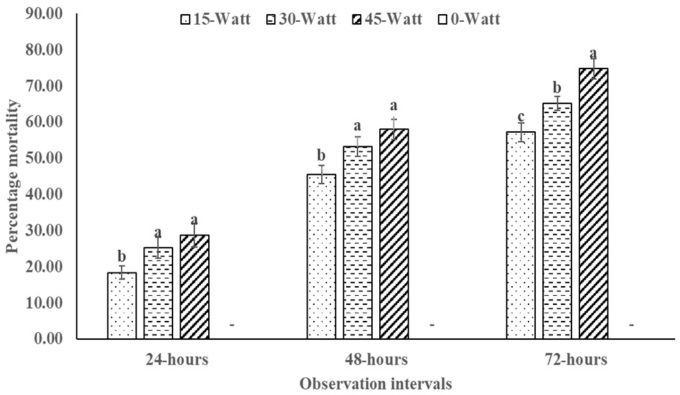

Figure 5: Overall mortality percentage of *Callosobruchus maculatus* adults treated with various powers of ultraviolet light at various observation intervals.
***Means followed by same letters are not significantly different from each other at individual observation interval; LSD values @ P < 0.05 24-h = 4.414; 48-h = 5.643; 72-h = 5.235**

The overall mortality percentage of *C. maculatus* adults treated with various powers of ultraviolet light at different exposure times over 24, 48, and 72 h is shown in Figure 5. A highly significant difference was observed in the effectiveness of different UV-light powers in causing mortality of adult *C. maculatus* after 24 h (F = 22.62, P < 0.001), 48 h (F = 8.20, P < 0.001), and 72 h (F = 8.60, P < 0.001). After 24 and 48 h, the highest adult mortality was recorded with 45-watt UV light treatment, at 28.80±3.30% and 58.00±2.80%, respectively, but these were not significantly different from mortality recorded with 30-watt UV light (25.20±2.90% and 53.20±2.70%, respectively). The lowest adult mortality at 24 and 48 h was observed with 15-watt UV light treatment, at 18.40±1.80% and 45.60±2.50%, respectively. Moreover, after 72 h, the highest mortality (74.80±2.80%) was recorded with 45-watt UV light, followed by 65.20±1.92% mortality with 30-watt UV light. The lowest mortality (57.20 2.60%) was observed with 15-watt UV light treatment.

The effect of various exposure timings of UV-lights on overall percentage mortality of adult *C. maculatus* is given in Figure 6. The results indicated a highly significant difference among the exposure timings to cause mortality of adults at 24-h (F = 66.75, P < 0.001), 48-h (F = 176.37, P < 0.001), and 72-h (F = 327.16, P < 0.001) of the application. After 24-h of the application of UV-light, the maximum adult mortality (38.70±4.00%) was recorded when different UV-light powers were applied for 5-h but the same was not significantly different from adult mortality recorded at 4-h exposure timing i.e., 32.70±2.80%. Moreover, the minimum mortality after 24-h of observation was recorded in 1-hour exposure treatment (12.00±1.40%), followed by the mortality recorded with 2-hour exposure of UV-lights (17.30±1.80%). Similar to 24-h, the highest percentage adult mortality after 48- and 72-h of the application was recorded at 5-h exposure time i.e., 62.70±3.20 and 76.00±3.60%, respectively, that was not significantly different percentage mortality of 59.30±3.40 and 71.30±2.70% after 48- and 72-h of observation, respectively observed at exposure time of 4-h. Overall, 1-hour exposure time of UV-light powers cause the lowest mortality of adult *C. maculatus* at various observation intervals.

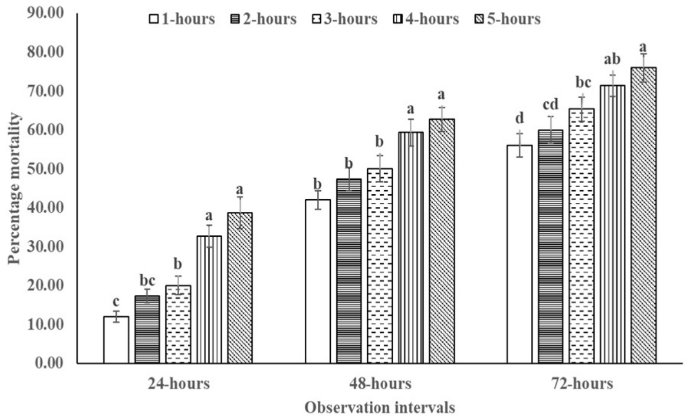

Figure 6: Overall mortality percentage of *Callosobruchus maculatus* due to various exposure timing of ultraviolet light radiations at various observation intervals.
***Means followed by same letters are not significantly different from each other at individual observation interval; LSD values @ P < 0.05; 24-h = 6.6141; 48-h = 8.4551; 72-h = 7.8441**

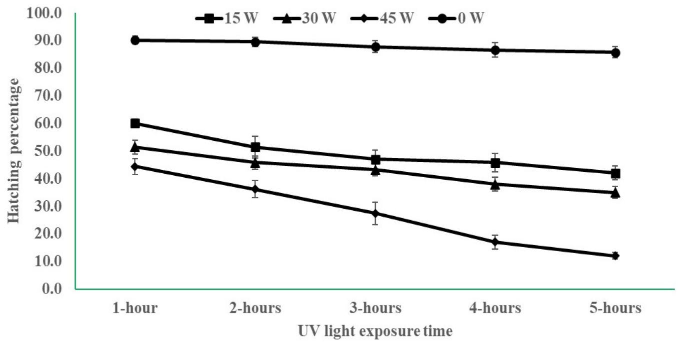

Figure 7: Effect of various exposure timings of ultraviolet light powers (watts) on hatching percentage of *Callosobruchus maculatus* eggs (LSD = 7.4419, P < 0.05).

The effect of Ultraviolet (UV) light on C. maculatus *eggs*

Figure 7 shows the results regarding the effect of various exposure timings of different UV-light power on the hatching success of *C. maculatus* eggs. A highly significant difference (F = 3.37, P = 0.005) was observed in UV-lights applied at various exposure timings on the hatching success of the eggs. Significantly the lowest hatching percentage of *C. maculatus* eggs (12.00±1.22%) was recorded at 5-h exposure to eggs with 45-watts of UV-light, followed by 17.00±2.55% hatching success, also recorded at 45-watt UV-light applied for 4-h. Moreover, the lowest hatching percentage recorded at 15- and

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30-watt UV-light was 42.00±2.55 and 35.00±2.24%, respectively both recorded at the exposure time of 5-h. The highest hatching success of *C. maculatus* eggs was recorded in control treatment when eggs were kept for one to five h in the rearing cage and ranged between 90.20±1.46 to 85.80±2.08.

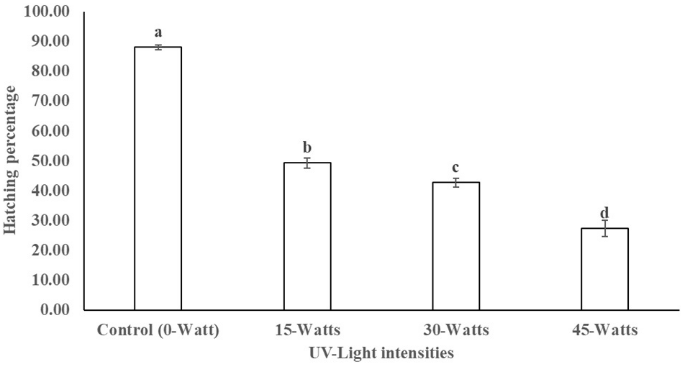

Figure 8: Impact of ultraviolet light powers on overall hatching percentage of *Callosobruchus maculatus* eggs.
***Means followed by same letters are not significantly different from each other at individual (LSD = 3.3281, P < 0.05).**

Results regarding the effect of various UV-power lights applied at different exposure timings on hatching success of *C. maculatus* eggs is given in Figure 8. A highly significant difference (F= 475.67, P < 0.001) in the effectiveness of UV-light powers was recorded on the hatching success of the eggs as significantly the lowest eggs were hatched at 45-watt UV-light (27.40±2.71%). Significantly the highest percentage of *C. maculatus* eggs hatched in control treatment (88.00±0.90%), whereas overall hatching percentage of eggs observed in 15- and 30-watt UV-light treatments was recorded as 49.24±179 and 42.68±1.53%, respectively.

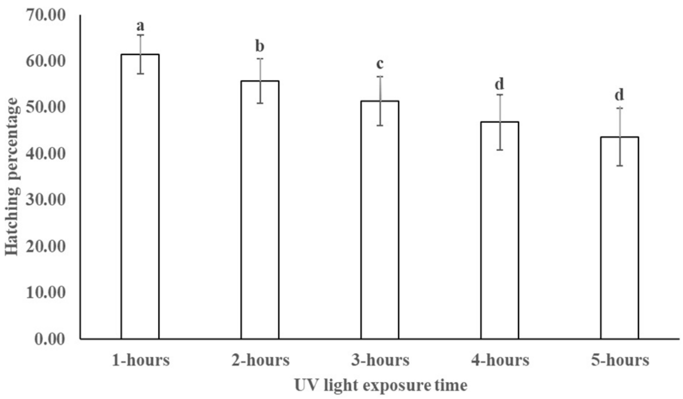

Figure 9: Impact of various exposure timings of ultraviolet light powers on overall hatching percentage of *Callosobruchus maculatus* eggs.
***Means followed by same letters are not significantly different from each other at individual (LSD = 3.7209, P < 0.05).**

Figure 9 illustrates results regarding the effect of various exposure times of UV-light powers on the hatching success of *C. maculatus* eggs. The results confirmed a highly significant (F = 28.60, P < 0.001) effect in the performance of various exposure timings of UV-light powers on hatching percentage of the eggs. Among treatments, significantly the highest hatching percentage of eggs (61.50±4.13%) was recorded at 1-hour exposure eggs with various UV-light powers, followed by 55.75±4.85 and 51.35±5.30%, recorded with 2- and 3-h exposure time. Moreover, exposure of *C. maculatus* with various UV-light powers for 5-h resulted in the lowest hatching success (43.70±6.20%), but the same was not significantly different from the hatching success recorded at 4-hour exposure time (46.85±5.93%).

Discussion

It was observed in the study that treatment of *C. maculatus* eggs and adults with various powers of UV-lights at different exposure timings exhibited a significant effect on their hatchability and mortality. A significant increase in the mortality rate of adult *C. maculatus* was observed when they were treated with increasing UV-light powers (15, 30, 45 watts) for extended times i.e., 1, 2, 3, 4, and 5-h. Accordingly, the highest overall mortality of adults (86.00±6.00%) was recorded at 45-watt power UV light applied for 5-h that was not significantly different from percentage mortality of 80.00±4.50% recorded in the same UV light power applied for 4-h. Moreover, maximum percentage adult mortality recorded in 30- and 15-watt UV light was 74.00±2.40 and 68.00±7.30%, respectively, both observed at 5-h exposure timing. Furthermore, application of UV-light was also found in reducing the hatchability of freshly laid *C. maculatus* eggs as higher reduction recorded with high power UV-lights and longer exposure timings.

It has been reported that by regulating various physical features i.e., temperature, humidity, heating, photoperiod, and pressure of the stored grain pest's environment can affect their growth, reproduction, and survival (Sahadia and Aziz, 2011). It is believed that most of the stored grains insect pests survive best and reproduce up to their maximum potential when they got optimum physical environment, especially temperature, and protein signaling phytochemical (Kalpna *et al*., 2022; Muhammad *et al*., 2023; Aritonang *et al*., 2024). Among these physical features, ultraviolet light has shown germicidal potential against various stages of insects, particularly adults and eggs (Arul *et al*., 2022). Thus, UV-lights are used for various purposes such as to disinfect the surfaces from insect eggs and attractant for adults (Bhardwaj *et al*., 2019; Faruki *et al*., 2005). Moreover, UV-radiations has also been proved to impact various biological parameters of the pulse beetles significantly (Heidari *et al*., 2016), besides other stored grains pests (Faruki *et al*., 2005). A recent study confirmed significant effect of UV radiation on various biological parameters of *C. chinensis* as compared to control as more than 60% reduction was recorded in fecundity, fertility, pupation, and adult emergence (Arul *et al*., 2022). Roy *et al*. (2017) also found significant impact of the application of short UV light (254 nm) as compared to freezing, silica gel, heating (60±1 °C), long UV light (366 nm), sodium sulfate and table salt against the *C. maculatus* on five common pulses (chana, mkabuli chana, moong, goat pea, and matar). Moreover, the three best treatments i.e., short UV-light, freezing and silica gel proved significantly better as compared to individual applications of the treatment with respect to grain damage of the stored pulses.

The effective results of UV-light against *C. maculatus* and *C. chinensis* in above-mentioned studies supported the findings of our study as significant mortality of *C. maculatus* was recorded in the UV-light treatments as compared to control, where no mortality was observed. Moreover, an increase in mortality and reduction in egg hatchability was recorded with increased exposure of *C. maculatus* adults and eggs with various UV-light powers. These findings were supported by the outcomes of Bhardwaj *et al*. (2019), who evaluated UV radiation (253.7 nm wavelength) against newly emerged adults and three-day old eggs of *C. maculatus* for different durations. It was observed that egg fertility and development of adults was significantly affected with increase in exposure duration of UV radiations. The longest mean generations time for both eggs and adults were recorded at 60 min exposure timing of UV radiation. However, Heidari *et al*. (2016) did not find any significant impact of UV-light applied at various exposure timings on the developmental time of various life stages of *C. maculatus*, but their application causes mortality of the immature stages. Moreover, they also observed significantly higher net reproductive rate in control (38.70 offspring) than 50 min exposure treatment of UV light (10.36 offspring). The intrinsic rate of increase and sex ratio of *C. maculatus* was also significantly affected by the UV lights applied at longer radiations i.e., 40 or 50 min than control treatment.

Modarres-Najafabadi *et al*. (2014) also studied the effect of various exposure timings of UV light irradiations on 1-, 2-, and 3-days old eggs. They found a decrease in the hatchability with increasing exposure time and age of the eggs as lowest hatchability (0.83%) was recorded in 3-days old eggs observed at 40-min exposure times, whereas 95% eggs were hatched in control. Results regarding other life table parameters i.e., net reproductive rate (R_0), intrinsic rate of increase (r_m) and finite rate of increase (λ) was lowest in 2-days old eggs treated with UV light radiations for 4-min, whereas highest of these parameters were recorded in control.

Other studies have also found that applying UV light against *C. maculatus* can significantly affect its various biological parameters, including its damage potential. Mahdi *et al*. (2015) discovered an inverse relationship between the LT50 values for UV radiation against *C. maculatus* and the exposure times, with a lower LT50 (1.35 h) observed when UV radiation was applied for 48 h. The adult-time mortality response of C. maculatus to 254 nm UV radiation was evaluated by Islam and Mustari (2010), who reported that the LT50 to kill 50% of the targeted adults was around 20 min. Additionally, the application of UV radiation significantly reduced fecundity in treated adults, while also increasing immature durations and adult emergence for both sexes. However, UV radiation was found to be less effective against the F1 generation of *C. maculatus* compared to the parental population.

Sedaghat *et al*. (2011) also found significant reduction in the hatchability of 1-,2-, and 3-days old eggs of *C. maculatus* when they were treated with UV-radiations (254nm) for 2, 4, 8, 16, 24, 32, and 40 min a comparatively high reduction in the hatchability was recorded when exposure time was increased. Moreover, significantly more effect of UV radiation was observed on older eggs of *C. maculatus* than 1-day old eggs. Similar trend of results was recorded by Singh and Das (2019) as they found significant reduction in adult emergence along with increased development time of *C. maculatus* when its eggs were treated with ultraviolet C (254nm) radiations for 2, 4, 6, and 8-min. Moreover, applying UV light did not negatively affect the grain quality and germination capacity.

An increasing trend in the mortality of adult *C. maculatus* with single exposure with various UV light powers over the exposure times of 24, 48, 72 h of observations that indicated that UV light has the potential to remain effective on the bodies of targeted insects and could be lethal later on. No previous studies were found to discuss the persistency of UV light, hence could be a potential area that could be exploited in future for better understanding of the mode of action of UV lights against insect pests.

Besides *C. maculatus,* UV radiations have also been used against various stored-grain pests such as *S. zeamais* (Ferreira *et al*., 2018; Tungjitwitayakul *et al*., 2016), *Trogoderma granarium* (Masoumi *et al*., 2021), *S. oryzae* and *T. castaneum* (Draz *et al*., 2021), *R. dominica* (Srivastava *et al*., 2019) and many others. All the studies recorded significant impact of UV radiation on mortality and reproductive parameters of these pests as mainly their effectiveness increased with higher exposure timings.

Conclusions and Recommendations

The application of UV-light (254nm) radiations (i.e., 15, 30, 45 watts) not only caused significant adult mortality in *C. maculataus* but also reduce hatching success of its eggs. The maximum adult mortality and lower egg hatchability of *C. maculataus* was recorded when exposed with 45-watt UV-light for a 4 or 5-h. Therefore, it is recommended that the use of 45-watt ultraviolet (254 nm) light in warehouses could reduce *C. maculatus population, hence resulted in lower losses to pulses*. However, further studies should be conducted on the evaluation of UV-light on large scale to determine its impact on various life stages of *C. maculatus* along with its cost effectiveness, both in small- and large-scale warehouses and godowns. It is also suggested to determine the effects of UV-lights on humans and other beneficial organisms to optimize its use against *C. maculatus* and other stored grains pests.

Declarations

Acknowledgements

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IRB approval

Not applicable

Ethical statement

Not applicable as no live animals involved in this study.

Generative AI and AI-assisted technology statement

No AI and AI-assisted technology used in preparation of this manuscript.

Statement of conflict of interest

The authors have declared no conflict of interest.

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