RESEARCH ARTICLE



# Effect of UV-A radiation as an environmental stress on the development, longevity, and reproduction of the oriental armyworm, Mythimna separata (Lepidoptera: Noctuidae)

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Abstract The ultraviolet light (UV-A) range of 320– 400 nm is widely used as light trap for insect pests. Present investigation was aimed to determine the effect of UV light-A radiation on development, adult longevity, reproduction, and development of  $F_1$  generation of Mythimna separata. Our results revealed that the mortality of the second instar larvae was higher than the third and fourth instar larvae after UV-A radiation. As the time of UV-A irradiation for pupae prolonged, the rate of adult emergence reduced. Along with the extension of radiation time decreased the longevity of adult females and males. However, the radiation exposure of 1 and 4 h/day increased fecundity of female adults, and a significant difference was observed in a 1 h/day group. The oviposition rates of female adults in all the treatments were significantly higher than the control. In addition, UV-A radiation treatments resulted in declined cumulative survival of  $F_1$ immature stages (eggs, larvae, and pupae). After exposure time of 4 and 7 h/day, the developmental periods of  $F_1$ larvae increased significantly, but no significant effects on  $F_1$  pupal period were recorded.

Keywords  $Mythimna$  separata  $\cdot$  UV-A radiation  $\cdot$ Development . Longevity . Reproduction

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## Introduction

Climate changes have versatile influences on consequences of abiotic stress, threatening the sustainability and productivity of agricultural systems. Abiotic and biotic stress factors have crucial role in insect life. There are numerous environmental stresses, e.g., depletion in ozone, increase in ultraviolet irradiation, loss of biodiversity and temperature, etc. UV radiation is a major environmental stress factor for many organisms (Meyer-Rochow [2000](#page-5-0); Schauen et al. [2007\)](#page-5-0). Currently, UV radiation is expecting a lot of considerations due to its possibly injurious effects on several organisms. UV proliferation can decline adult longevity, enhance oviposition rate, and reduce fecundity and fertility (Faruki et al. [2005](#page-4-0); Zhang et al. [2011\)](#page-5-0). UV stimulates ROS, which can spoil nucleic acid, membrane lipids, and proteins (Vile and Tyrrell [1995](#page-5-0)). These varieties of damages in above molecules cause substantial effects on biology such as genetic variation, cytotoxicity, and transformation in signaling cell pathways (McMillan et al. [2008\)](#page-5-0).

The oriental armyworm, Mythimna separata (Lepidoptera: Noctuidae), is an important migratory pest in eastern Asia. It has caused damage to wheat, corn, and rice crops since 1970 in China (Yang et al. [1984\)](#page-5-0). Its seasonal migration has been studied in China (Chen and Bao [1987](#page-4-0); Chen et al. [1995\)](#page-4-0). During winter, this migratory pest settled in southwestern China and flies to northern China to damage summer crops. Recently, the invasion of armyworm has caused damage to crops; especially corn was damaged in neighboring Liaoning, Hebei, and the Inner Mongolia province (Ye [2012\)](#page-5-0). The moths of nocturnal insects are frequently drawing intention to UV light due to their innate behavior which can be a base for their control (Wei et al. [2000;](#page-5-0) Emura and Tazawa [2004\)](#page-4-0). UV radiation disturbs an insect's fitness by limiting its ability to find a native mate, its reproduction by affecting its longevity, or disturbing the harmony of its circadian rhythms

<span id="page-1-0"></span>(Barlett et al. [1968;](#page-4-0) North and Holt [1968;](#page-5-0) Proverbs [1969;](#page-5-0) Van Steenyk et al. [1979;](#page-5-0) LaChance et al. [1975](#page-4-0); Tsubaki and Bunroongsook [1990;](#page-5-0) Hasan [1995\)](#page-4-0). UV light broadly used as integrated pest management (IPM) to control different kinds of insect pest populations such as lepidopteron (Antignus [2000;](#page-4-0) Steinbauer [2003](#page-5-0); Kojima et al. [2005](#page-4-0)). UV radiation directly affects the insect behavior, developmental physiology, and biochemistry (Gunn [1998;](#page-4-0) Mackerness et al. [1999;](#page-4-0) Mazza et al. [2002](#page-4-0)). Various studied discovered that immature developmental stages are more susceptible to indirect and direct UV radiation than mature insects (Watters [1976](#page-5-0); Tilton and Brower [1983;](#page-5-0) Shayesteh and Barthakur [1996;](#page-5-0) Zhao et al. [2007](#page-5-0); Zhang et al. [2011](#page-5-0)), and it is also reported that UV radiation effect on living organisms is associated to cellular development. Low intensity of UV-A radiation increased the lifespan of Drosophila, and the high intensity of UV-A radiation can lead to prolonged life expectancy shortened (Zheng [2011](#page-5-0)). In the present study, we examined UV-A irradiation effect on survival, reproduction, different development stages, and as well as  $F_1$  generation of M. separata. The purpose of the present study is to estimate the effect of UV-A radiation on physiology of M. separata.

#### Materials and methods

## Experimental insects

*M. separata* were reared at the room temperature  $26 \pm 2^{\degree} \text{C}, 60$  $\pm 10$  % RH, and 14:10 h L/D. An artificial diet was used to feed the larvae as mentioned in Chun [\(1981\)](#page-4-0).

#### Larvae and pupae

The second, third, and fourth instar larvae and 48-h-old pupae were collected from the food medium kept in separate petri dishes. The UV-A radiation was given to larvae and pupae 1 h after the start of scotophase. The irradiation dose was approximately at 350 W/cm<sup>2</sup>. Larvae and pupae were exposed to UV-A for 0, 4, 8, and 16 min. The larval mortality was assessed up to pupation. There were 50 larvae and 30 pupae per treatment,

Fig. 1 UV-A radiation effect on different instars of Mythimna separata larvae. Values (mean ± SEM) followed by different letters within a column are significantly different by Tukey HSD test  $(P < 0.05)$ 

with three replications. Irradiated pupae were kept in plastic containers for adult emergence.

## Adult longevity and reproduction

UV light-A (X-series, peak emission 365 nm; Spectronics, Westbury, NY) were used in the experiment. Male and female adults were exposed to UV-A for the different period of times, i.e., 0 (control), 1, 4, and 7 h (hours) per day, until the death of males and females. Ten female–male pairs were used in each treatment with three replications of UV-A radiation. Each female–male pair was reared in a single plastic cup (10 cm 7 diameter) was tested. The plastic cups were closed with cotton net yarn and kept butter paper in glass for egg laying. Cotton net yarn and batter paper were replaced daily. The fecundity, ovipostion rate, and survival of male and female were recorded on the daily basis. The experiment was terminated until the males and females died.

#### Development of the  $F_1$  generation

Eggs laid by adults of the P generation were referred to as the  $F_1$  generation. Cotton yarns with eggs were sprinkled with water and then transferred to a separate plastic petri dish (7 cm diameter). One hundred fresh eggs were sampled randomly and incubated into three replications (per treatment 300 eggs) in plastic petri dishes. To estimate the hatching percentage of eggs, the newly hatched larvae were noted daily. A plastic container  $(3 \times 3 \times 4$  cm) was used to rear 60 newly hatched larvae individually in three replications per treatment (total 180 larvae) to observe the larval period. The number of pupa and the timing of pupation were recorded, and 40 newly obtained pupae were transferred into new separate plastic containers with three replications per treatment (total 120 pupae). The number of the adult and timing of the emergence was logged. The experiments were performed at  $26 \pm 2$  °C, 60  $\pm$  10 % RH, and 14:10 h L/D photoperiod.

#### Statistical analysis

The software SPSS (SPSS Inc., Chicago, IL, USA) was used for data analysis. The one-way analysis of variance (ANOVA)





Fig. 2 Adult emergences (%) of *Mythimna separata* from UV-irradiated pupae. Values (mean  $\pm$  SEM) followed by different letters within a column are significantly different by Tukey HSD test ( $P < 0.05$ )

was used for analysis of data, and means were compared Tukey HSD test at  $P < 0.05$ . Data of egg hatch, survival of larvae, and pupae were arcsine. Square-root transformed prior to analysis.

## **Results**

# Effect of UV-A radiation on larval mortality and adult emergence

The larval mortalities of second, third, and fourth instars gradually increased as UV-A radiation exposure time increased. The larval mortality of second and third instars significantly increased  $(F = 132.85, P < 0.001, df = 3.8; F = 45.60,$  $P < 0.001$ , df=3.8), respectively, at the exposure period of 4, 8, and 16 min UV light. It was observed that third instar larval mortality was significantly higher than fourth instar larvae  $(F= 32.66, P< 0.001, df= 3.8)$ . A high mortality rate was observed in fourth instar larvae at the longest exposure time of 16 min. No significant deference was observed between control and 4-min UV irradiation. We found that earlier larval stage (second instars) were more sensitive to UV at any level of mortality counted than third and fourth instar larvae (Fig. [1](#page-1-0)).

Adult emergence of UV-A-exposed M. separata pupae significantly reduced at 8 and 16 min  $(F=30.364 \ P<0.001$ ,  $df = 3.8$ ). There was no significant difference between control and 4-min UV light exposure (Fig. 2).

#### Effect of UV-A radiation on adult longevity

A significant decrease in female longevity was observed when female adults were exposed to UV-A radiation for 1, 4, and 7 h/day ( $F = 50.261$ ,  $P < 0.001$ , df = 3.8). There was a significant decrease in male longevity found when male adults were expose to UV-A radiation for 7 h/day ( $F = 6.025$ ,  $P < 0.01$ ,  $df = 3.8$ ), while no significant difference was seen in male longevity between 1 and 4 h/day UV radiation as compared to control group (Fig. 3a, b).



Fig. 3 Effect of UV-A radiation on adult longevity of Mythimna separata, for a females, b males. Error bars indicate SEM. Bars with different letters are significantly different by Tukey HSD test ( $P < 0.05$ )

#### Effect of UV-A radiation on reproduction

UV-A radiation effect on reproduction of M. separata from the exposure of 1 and 4 h/day resulted in a fecundity increase as compared to control. We found a significant difference after the exposure of 1 h/day  $(F=6.208, P<0.017, df=3.8)$ . Females laid fewer eggs compared to control in the oviposition period, but statistically, the difference was not significant when they were exposed to UV-A radiation for 7 h/day. Oviposition rate was significantly affected by UV-A radiation  $(F= 11.089, P< 0.003, df= 3.8)$ . The rate of oviposition was increased as the exposure time increased, and it reached to a maximum at 7 h/day as compared to all other groups (Table 1).

# Effect of the UV-A radiation on development of the  $F_1$ generation

Data on the effect of UV-A radiation indicated a significant decrease on 7 h/day in F<sub>1</sub> eggs ( $F = 6.606$ ,  $P < 0.01$ , df=3.8),

Table 1 Effect of UV-A radiation on reproduction of Mythimna separata

Exposure time $(h/day)$	Fecundity (eggs female)	Oviposition rate (eggs/female/day)
$\Omega$	$840.73 \pm 17.93b$	$76.62 \pm 1.16b$
$\mathbf{1}$	$903.60 \pm 17.66a$	$95.50 \pm 0.51a$
$\overline{4}$	$851.36 \pm 15.58$ ab	$96.72 \pm 4.61a$
7	$813.63 \pm 6.20$	$101.35 \pm 4.47a$

Values (mean  $\pm$  SEM) followed by different letters within a column are significantly different by Tukey HSD test  $(P < 0.05)$ 

and it was not significantly different for the survival of  $F_1$ larval ( $F = 2.528$ ,  $P = 0.13$ , df = 3.8). A significant decrease was observed on 4 h/day in  $F_1$  pupal survival ( $F= 10.377$ ,  $P < 0.004$ , df=3.8). UV-A radiation exposure resulted a significant reduction in cumulative survival as compared to the control group for a period of 1, 4, and 7 h/day (Table 2).

The larval and pupal developmental periods of  $F_1$ M. separata was also affected by UV-A radiation. It was found that  $F_1$  larval developmental period increased in contrast to control, as the exposure of UV-A radiation was given to adults. At the duration of 4 and 7 h/day, a significant  $(F= 39.787, P< 0.001, df= 3.8)$  interruption was seen in the development of  $F_1$  larvae. The larvae of  $F_1$  took longest time to attain the pupal stage for the longest UV-A exposure time (7 h/day). There was no significant ( $F = 0.854$ ,  $P = 0.50$ ,  $df = 3.8$ ) effect seen on the developmental period of  $F_1$  pupae. The emergence time for the adult stage for both  $F_1$  pupae in the control and all the treated groups were same (Table 3).

## Discussion

UV-A radiation has a strong capability to affect the physiology of insects. Our results showed that UV-A radiation has more harmful effect on younger larvae as compared to older larvae. Similar results were also reported by (Qureshi et al. [1970](#page-5-0); Faruki et al. [2005\)](#page-4-0). Mortality of the irradiated larvae was directly related to the dose of radiation, i.e., mortality gradually increased as the dose of radiation increased. Recently, it is found that larval mortality was completely associated with radiation doses (Pandır and Guven [2014](#page-5-0); Guven et al. [2015\)](#page-4-0). Insect mortality ratio enhances with increasing UV irradiation time. It may be due to the protein and fat level in the insect body. These molecules are less in earlier larval instar and higher in later instar. These molecules may have the role in immune defense system, so they hinder the UV light effect in later stages. Therefore, mortality is higher in earlier larval instar as compared to later instars. Adult emergence of UVexposed M. separata pupae decreased as the time of irradiation increased. A significant reduction in adult emergence from irradiated pupae was observed (Hasan et al. [1998](#page-4-0);

Table 3 Effects of UV-A radiation on developmental periods of larvae and pupae of  $F_1$  Mythimna separata

Exposure time (h/time)	Larva days	Pupa days
$\theta$	$18.56 \pm 0.11c$	$9.54 \pm 0.23a$
1	$18.63 \pm 0.13c$	$9.63 \pm 0.43a$
$\overline{4}$	$19.12 \pm 0.05$	$9.74 \pm 0.22a$
7	$19.85 \pm 0.03a$	$10.11 \pm 0.02a$

Values (mean  $\pm$  SEM) followed by different letters within a column are significantly different by Tukey HSD test  $(P < 0.05)$ 

Faruki et al. [2007\)](#page-4-0). In our results, longer exposure time highly affected pupal stage. The possible reason is that pupal stage is static and UV irradiation directly enters and disturbs key physiologically processes (Tilton and Brower [1983\)](#page-5-0).

Meng et al. [\(2009\)](#page-5-0) reported that ultraviolet light could induce oxidative stress in adult insects. These radiations can upset the working capacity of proteins and augment the resultant product of protein oxidation in adult of M. separata. These processes decreased longevity and enhanced the oviposition rate (Zhang et al. [2011\)](#page-5-0). Similar results were found in the present study in which the adult longevity was shortened, and rate of oviposition was elevated in M. separata. UV-A radiation extremely affected the biological parameters of P generation. Fecundity was increased after 1 and 4 h/day UV-A radiation as compared to control groups. But, significant difference was noted after 1 h/day groups. Fecundity was lesser than control at longer exposure time 7 h/day, but there was no significant difference. The oviposition rate of female adult in all UV-A radiation treatment groups were significantly increased as compared to control groups. After UV-A radiation, why fecundity and oviposition rate was increased remains unknown. Reason could be as the theoretically life history forecasts that some traits may exchange one another, particularly when each trait is vigorously expensive (Zera and Harshman [2001](#page-5-0); Zera and Denno [1997\)](#page-5-0). In common life history theory, it is a crucial prophecy that reproduction sustains a survival cost (Williams [1966;](#page-5-0) Roff [1992\)](#page-5-0). Consequently, we presume that there is an exchange in energy sharing between the longevity and reproduction of M. separata adults under

Table 2 Effects of UV-A radiation on egg hatch and survival of immature stages of  $F_1$ Mythimna separata



Values (mean ± SEM) followed by the same letter within a column are not significantly different by Tukey HSD test ( $P < 0.05$ ). The total numbers of insects in each treatment were 300, 180, and 120 eggs hatch, larval, and pupal survival, respectively

<span id="page-4-0"></span>UV stress. Previously, it was known in lepidopteron moths that they have the ability to enhance their antioxidant defense system in response to toxic effects of ultraviolet radiation (Meng et al. [2009\)](#page-5-0). It seems that this reaction is possibly expensive for the organism in terms of energy. In response to UV-A exposure, reproductive costs and antioxidant defenses are developed, so life history theory forecasts that existence capability must be faded to a certain limit (Holloway et al. 1990; de Jong and van Noordwijk 1992), and it was seen in the reduction of longevity in M. separata adult. In our experimental condition, UV-A radiation on hatched eggs and pupae survival of  $F_1$  generation significantly differed while there was no significant difference observed in larval survival. Present findings indicate that cumulative survival of  $F_1$  generation control was comparatively higher than UV-A radiation-treated groups. We observed that UV radiation disturbs the  $F_1$  generation, and longer time UV radiation can reduce the cumulative survival of  $F_1$  immature stages. The results revealed that significant difference was not found in developmental periods of  $F_1$  pupae, while significant change was seen in the developmental periods of  $F_1$  larvae between UV-A exposure and control insects.  $F_1$  larvae of UV-A-treated adults took longer time to become pupae. This result revealed that UV-A radiation has harmful effects on the larval development period of  $F_1$  generation. UV radiation can be used as an ecological stress factor in insects Meng et al. ([2009](#page-5-0), [2010](#page-5-0)). Many insects have the ability to prolong their developmental stages to prevent themselves from adverse effects of environmental conditions for their survival, which is the strategy of organisms to compensate themselves according to the environmental conditions for the protection of harmful effect on growth (Gintenreiter et al. 1993; van Ooik et al. [2007](#page-5-0)).

## Conclusion

The present investigation revealed that UV-A radiation had high effect on survival, reproduction, different development stages, and  $F_1$  generation of M. separata. Earlier instar larvae of M. separata are more sensitive to UV radiation than later instar larvae. Adult emergence of UV-A-exposed M. separata pupae reduced as the time of irradiation increased. UV radiation reduces adult longevity and increases fecundity and oviposition rate of M. separata. It also affected the developmental stage of M. separata  $F_1$  generation.

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