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EFFICACY OF GAMMA RADIATION ON THE GREATER WAX MOTH *GALLERIA MELLONELLA* L.

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ABSTRACT

The effect of ionizing radiation from 50 to 450 Gy on the eggs and larvae 100 to 1500 Gy. on larvae of greater wax moth *Galleria mellonella* L. The results revealed that the egg hatchability reduced to 50% at 102.70 Gy. In the most radiation resistant eggs, 350 Gy resulted in no pupal formation while a dose of 250 Gy resulted in no adult formation. Within 51.53 hr, a dose of 500 Gy reduced the larval survival to 50%. With 148, 574 and 680 Gy, the proportion of larvae that matured into adults reduced to 50%, 10%, and 0%, respectively. A radiation dose between 300 and 400 Gy was found adequate for egg sterilization and larval mortality.

Key words: *Galleria mellonella, g*amma radiation, lethal dose, radiation dose, 50-1500 Gy, egg hatchability, pupation, adult formation, egg sterlization, larval mortality

The greater wax moth (*Galleria mellonella* L.) is a well-known pest that harms honey bee colonies. It causes significant economic losses of 60-70% to Indian beekeepers (Hanumanth et al*.,* 2009). Roughly US$ 4.5 million dollars is lost in United States, with about US$ 1 million occurring in Florida alone (Kondrateva et al*.,* 2020). In tropical and subtropical areas, *G. mellonella* is the limiting factor that severely harm honey bee populations (Charles et al., 2017), it occurs due to various reasons, viz; poor nutrition, illness, the loss of the queen, or widespread pesticide poisoning (Pirk et al., 2015). *Galleria mellonella* also contributes to the spread of infectious illnesses, including the foulbrood (Goulson et al., 2015). Management of this pest has drawn significant interest (Shimanuki et al., 1980; Williams, 1997). *Galleria mellonella* has historically been subjected to heat and cold treatments to end all stages of its existence (Bombelli, 2017; Charles et al., 2017). Chemical pesticides used detrimental side have effects on non-target organisms and preradiation is used to combat this problem (Hallman and Blackburn, 2016). A dose of 400 Gy phytosanitary irradiation is necessary to destroy wax moth eggs (Mansour, 2020). However, significant research on the dose required to kill the larvae has not been quantified.

In the current study, an effort was made to apply gamma radiation to combat *G. mellonella* with a view to generate baseline data calculating for LD_{50}) values for the eggs and larvae. This study looked at several variables, including how gamma radiation affects egg hatching, pupation, and adult emergence. Additionally, the effects of gamma radiation on the irradiated larvae that develop into pupae and adult stages were evaluated, as well as the fluctuation in mortality over time for the larvae exposed to various dosage levels.

MATERIALS AND METHODS

The Department of Nematology at CCSU in Meerut, Uttar Pradesh, provided the *G. mellonella* culture. The culture was maintained in a growth chamber with controlled humidity and temperature. The culture was raised on an artificial diet (Firacative et al., 2020). Eggs and larvae of *G. mellonella* were collected by the standard procedure (Mansour, 2020). The age of the eggs was noted from the first day till egg hatch. Different age groups' of eggs and larvae received varying levels of radiation. A gamma irradiation chamber (model Gamma Cell Elite-I) with a Cs-137 radioactive source was used. Paper cups containing eggs attached to paper strips were put into the irradiation chamber, while larvae were directly introduced into the paper cups. The eggs were returned to the BOD incubator and reared on diet medium. Within two weeks, some of these eggs hatched and became larvae. After five weeks, some larvae began to weave cocoons around themselves to develop into pupae. To produce adults, the pupae were housed in sterile containers. Adult food in the form of a 10% sucrose solution was provided. Radiation doses used ranging from 50 to 450 Gy with an incremental dose of 50 Gy were administered to eggs of various age

groups (1-6 days old). For the following 10 days, the irradiated eggs were watched to count hatching eggs. A mixed population of larvae was separately exposed to radiation at doses ranging from 100 to 1500 Gy with a 100 Gy increment. To help with the computation of the LD_{50} dose, which kills 50% of the exposed larvae within 24 hr, the irradiated larvae were observed for the mortality. Four samples of 50 each, were used for each dose. A mixed population of larval instars was selected to reflect the real scenario of an infected comb. To determine the LD_{50} dose with the larvae subjected to doses of 500, 1000 and 1500 Gy were observed for the following 96 hr. The % mortality was documented every 24 hr. Data on the mortality and the sterility of irradiated eggs were subjected to probit analysis using IBM's SPSS software. The same software was used to perform Pearson's chi-square test to determine the significance of the relationship between dosage and mortality. Kolmogorov-Smirnov and Shapiro-Wilk tests were used to determine the data's normality.

RESULTS AND DISCUSSION

It was observed that the *G. mellonella* eggs are more radiosensitive while they are young and eventually become resistant as they age, but, as the exposure increased, the resistance reduced (Fig. 1). For eggs that were irradiated at 1-2, 3-4, and 5-6 days of age, respectively, the regression results demonstrated a substantial inverse relationship between gamma radiation dose and egg hatch (P 0.05, $R^2 = 0.916, 0.877$, and 0.954). The most radio-resistant eggs were discovered to be 5-6 days old, whereas the radio-sensitive eggs were found to be 1-2 days old. A dose of 250 Gy resulted in 0% egg hatch in the 1-2 days old eggs and reduced the egg hatch to 12.75 and 29.50%, respectively in 3-4 and 5-6 days old eggs. The egg hatch reduced significantly with dose (Table 1). A dose of 50Gy affected the survival of larvae ($p < 0.05$) in most radio-resistant eggs (5-6 days old eggs)

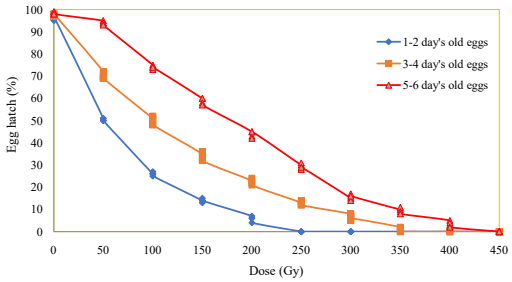

Fig. 1. Effect of ionizing radiation on % egg hatch of *Galleria mellonella*

and a dose of 350Gy completely arrested the survival of larvae. For the age groups of 1-2, 3-4, and 5-6-day-old eggs, respectively, a strong negative connection between the dose and survival to adult stage was found ($p < 0.05$, $R^2 = 0.979, 0.988, 0.906$). For a mixed population of the larval instars, it was observed that a dose of 350Gy reduced the irradiated larvae becoming pupae to 50%. Doses of 148, 574 and 680Gy reduced the irradiated larvae turning into adults to 50, 10 and 0%, respectively ($p < 0.05$, $R^2 = 0.698$) (Fig. 2). The graph clearly shows a relationship between the proportion of mortality over time and radiation exposure. The mortality of larvae exposed to doses of 500, 1000 and 1500 Gy was 21.72, 41.63 and 63.98%, respectively, at 24 hr. A dose of 500 Gy caused 83% larval mortality after 96 hr and lowered the survival to 50% after 51.53 hr.

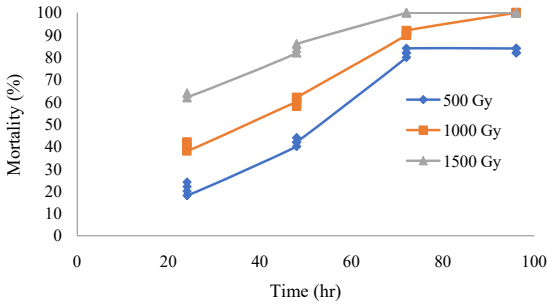

Fig. 2. Effect of ionizing radiation on cumulative larval mortality in *Galleria mellonella*

Table 1. Variation of survival with dose on the age of the irradiated eggs of *Galleria mellonella*

Dose (Gy)	1-2 days old eggs			3-4 days old eggs			5-6 days old eggs		
	Survival to larval stage	Survival to pupal stage	Survival to adult stage	Survival to larval stage	Survival to pupal stage	Survival to adult stage	Survival to larval stage	Survival to pupal stage	Survival to adult stage
50	50.50%	41.25%	39.00%	70.00%	60.25%	56.50%	93.75%	80.50%	73.75%
100	26 .00%	24.50%	22.75%	50.00%	42.50%	39.25%	74.00%	60.50%	53.75%
150	14.00%	15.75%	8.75%	34.00%	28.25%	25.00%	58.75%	36.75%	29.75%
200	5.75 %	8.75%	0.00%	22.50%	11.50%	8.75%	43.75%	23.75%	16.75%
250	0.00%	0.00%	0.00%	12.75%	5.00%	0.00%	29.50%	13.50%	8.50%
300	0.00%	0.00%	0.00%	6.50%	0.00%	0.00%	15.50%	3.25%	1.50%
350	0.00%	0.00%	0.00%	0.75%	0.00%	0.00%	9.50%	0.00%	0.00%
400	0.00%	0.00%	0.00%	0.00%	0.00%	0.00%	3.50%	0.00%	0.00%
450	0.00%	0.00%	0.00%	0.00%	0.00%	0.00%	0.00%	0.00%	0.00%

Use of ionizing radiation for pest disinfestations started in the early 20th century (Runner, 1916). Phytosanitary irradiation of various insect pests with a dose ranging from 150 to 400 Gy is being used extensively (Follett et al., 2022). The present results indicate a dependence on the dose. These findings are similar to those of Nadel et al. (2018), Jafari et al. (2010) reported that the most effective dose for the sterilization of the male pupae of *G. mellonella* as 350 Gy. The present results are in close agreement with those of Hallman et al. (2010); Mansour (2020); Ayvare in contrast to those of Milcheva az et al. (2008); Mansour and Al-Attar (2012) on many lepidopteran pests. The observation on doses for different parameters are close to that of Mansour (2010; 2015; 2016). The LD_{50} observed now are in contrast to those of Milcheva (2004) and it might be due to various reasons (Hallman, 2000; White et al., 1977). Essentially, ionizing gamma radiation has quarantine potential for the management of the *G. mellonella* and a dose of 300 to 400 Gy is quite satisfactory.

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