

**INDUCTION OF STERILITY IN THE UZI FLY, *EXORISTA BOMBYCIS* (LOUIS)
(DIPTERA : TACHINIDAE) THROUGH GAMMA IRRADIATION OF PUPAE**

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Four, five, six and seven day old male and female pupae of *Exorista bombycis* were subjected to a predetermined dose of 2 Kr gamma radiation. Adult emergence declined significantly in 4 and 5-day old irradiated pupae compared to control. Average life span of adults emerging from 5 day-old irradiated pupae decreased significantly when compared with control. There was no significant variation in the period of survival between the male and female flies emerging from normal and irradiated pupae. Fecundity of normal females mated with sterile males emerged from irradiated pupae was not significantly different from control, but the same in females emerging from 5, 6 and 7-day old irradiated pupae was significantly lower when they mated with normal males. No egg laying was recorded in the females emerging from 5 and 6 day-old irradiated pupae as the ovarian development was completely inhibited. The radiation dose (2 Kr), which induced total sterility in males and females, did not affect the mating ability of males and receptivity of females. Under field release programme of the irradiated flies in a limited area, a ratio of 10:1:1 (sterile male:normal male:normal female) was found effective in suppressing the progeny of *E. bombycis*.

Key words : Gamma irradiation, *Exorista bombycis*, sterility, mating competitiveness.

INTRODUCTION

The uzi fly, *Exorista bombycis*, is a serious endoparasitoid of the silkworm, *Bombyx mori*. The parasitization process commences with the young parasitoid maggot hatching from the egg laid on the host larval body feeding on various tissues after piercing through the host integument. The parasitisation process culminates in the death of the host in either larval or pupal stage depending on the host stage parasitised. As a result, 10-20% loss to silkworm crops is experienced by the farmers in the southern silk producing states of Karnataka, Andhra Pradesh and Tamil Nadu (Kumar *et al.*, 1993), which is far in excess of the economic injury level.

Sterility in *E. bombycis* when its adults were fed tepa and thiotepa was reported (Singh & Mukherjee, 1973; Datta & Mukherjee, 1978). Kumar *et al.* (1990) have reported that gamma radiation induced sterility in both the sexes of uzi fly when the pupae were irradiated. Gamma radiation is known to induce sterility in males of several dipterans viz. the screw-worm fly, *Cochliomyia hominivorax* (Knippling, 1960), the house fly, *Musca domestica* (Schmidt *et al.*, 1964; Kilgore & Douth, 1967) and the Mediterranean fruit fly, *Ceratitis capitata* (Katiyar & Ramirez, 1970; Zumreoglu *et al.*, 1979).

Considering the economic importance of *E. bombycis*, an attempt has been made in the present investigation to record the influence of gamma radiation on adult emergence, longevity, reproductive potential and mating competitiveness of the flies emerging from the irradiated pupae. The outcome of the work helps to formulate strategies to manage uzi fly based on the principles of autocidal control.

MATERIALS AND METHODS

Culturing of uzi fly : The uzi pupae (4-7 day-old) used in the present investigation were obtained from the laboratory stock culture maintained in the Pest Management Laboratory of the Central Sericultural Research and Training Institute (CSRTI), Mysore. The parasitoid culture was maintained on the larvae of *B. mori* at 24-27°C temperature and 65-85 % relative humidity.

Source and dose of gamma radiation : Four to seven day old pupae of uzi fly were irradiated in a gamma chamber-4000 containing ^{60}Co as irradiator at a radiation emission rate of 0.07mega r/h at CSRTI, Mysore. A dose of 2 Kr which was reported to induce sterility in *E. bombycis* (Kumar *et al.*, 1992) was used for exposing the pupae. Each treatment comprised 3 replications of 100 pupae each. Untreated pupae of corresponding ages served as control.

Observations : The irradiated pupae were placed separately in nylon netted oviposition cages (30 x 30 x 30 cm) with a long sleeve on one side and observed the emergence and longevity of adults. The emerging adults were provided with sucrose cubes as food and soaked absorbent cotton as source of water. To record the effect of gamma radiation on the fecundity and fertility, the following crosses were set up between the normal and irradiated adults:

- Sterile male x normal female.
- Sterile female x normal male
- Normal female x normal male (Control)

Ten sets each with a pair of fly were maintained in oviposition cages. Fourth and fifth instar silkworm larvae were provided for oviposition. New set of larvae was provided daily for egg laying. Observations were made daily. Appearance of black scar(s) which is the typical symptom of parasitisation by uzi fly on the integument of host larvae was considered as an indication of egg hatch. The experiment was repeated thrice.

Brood test : Brood test in irradiated flies was conducted following the methods of Datta & Mukherjee (1978).

Mating competitiveness : To study the mating ability of males emerging from irradiated pupae of *E. bombycis* vs normal males under laboratory conditions, the irradiated males were caged with normal males and females in different ratios from 0:1:1 to 10:1:1 (Sterile males : normal males : normal females). Normal flies of both the sexes of the same age were also maintained from the untreated lot to serve as control. Data derived from various observations were subjected to Analysis of Variance after Gomez & Gomez (1984).

RESULTS AND DISCUSSION

Effect on adult emergence

Adult emergence from 4, 5, 6 and 7 day-old irradiated pupae was 3.0, 69.0, 86.0 and 87.33 %, respectively (Table I). A significant decline ($P < 0.05$) in adult emergence was recorded in 4 and 5 day-old irradiated pupae, but no such reduction in adult emergence was observed in 6 and 7 day-old irradiated pupae when compared with control (90.20 %). These findings are in agreement with the results obtained with the black blowfly, *Phormia regina* (Bushland & Streeter, 1971) and differ from those recorded with *C. capitata* (Zumreoglu *et al.*, 1979) and *C. hominivorax* (Bushland & Hopkins, 1953) where the test insects exhibited relatively higher tolerance to gamma radiation (5 day-old pupae with 2.5 Kr). The variation in tolerance to radiation exhibited by these insects seems to be an insect species associated trait.

The pupation period (12 days) was found similar in both the normal and irradiated pupae of *E. bombycis*.

Effect on longevity

Longevity of both male (12.2 days) and female flies (13.0 days) emerging from 5 day-old irradiated pupae decreased significantly ($P < 0.05$) when compared with control (14.7 and 15.5 days for male and female, respectively). However, the average life span in male (13.8 days) and female (14 days) flies emerging from the 6 and 7 day-old treated pupae was not significantly lower than control.

Like *E. bombycis*, the adults emerging from gamma irradiated pupae of *C. hominivorax* (5 day-old) with 2.5-5.0 Kr (Bushland & Hopkins, 1953) and *Dacus cucurbitae* exposure of pupae 24 h before adult eclosion) with 10.0-10.5 Kr (Steiner *et al.*, 1965) have exhibited an appreciable decline in their longevity. By contrast, *C. capitata* exhibited higher tolerance when exposed to 10.0-12.5 Kr gamma radiation, where no deleterious effect on the longevity of test insect was observed (Katiyar & Velerio, 1963).

Effect on fecundity

Due to poor emergence of the adults from 4 day-old treated pupae, studies on the fecundity and fertility on these flies were not conducted. Further, only those treatments which have shown higher adult emergence and significant decline in fertility compared to control have been presented for discussion.

a) Sterile male x normal female: The average fecundity recorded for crosses involving treated male and normal female flies emerging from 5, 6 and 7 day old pupae was 413.3 ± 67.4 , 456.3 ± 69.20 and 418.6 ± 19.2 , respectively as against 485.6 ± 56.2 in the control (Table I). Though the fecundity of the flies in the treated batches declined, it was not significantly lower than that in control.

b) Sterile female x normal male: Female flies emerging from 5 and 6 day-old irradiated pupae when mated with normal males failed to oviposit. Such females showed highly

reduced ovarian development. Though the flies emerging from 7 day-old treated pupa did oviposit, there was a significant decline in fecundity (29.3 ± 1.5) in these flies. Mating was found normal in all the above cases.

Results pertaining to decline in fecundity when normal females mated with irradiated males are in close conformity with the findings of Bushland & Streeter (1971) who exposed the pupae of *Phormia regina* to 2 Kr gamma radiation.

Kumar *et al.* (1990) have reported a highly retarded ovarian development following the exposure of *E. bombycis* pupae (8 days-old) to 8 Kr gamma radiation as observed in the present study where the pupae (5-7 day-old) were exposed to only 2 Kr gamma radiation. As a result, the irradiated females failed to lay eggs though their longevity was comparable with those of control batches, except the flies emerging from 5 day old irradiated pupae.

Effect on fertility

a) Irradiated male x normal female : Though the fecundity of the normal females mated with irradiated males did not vary significantly, the irradiation adversely affected the fertility in all the treatments (Table I). However, a fertility of 22.70 ± 2.12 % was recorded in the flies emerging from 7 day-old irradiated pupae. No egg laying was observed in flies emerging from 5 and 6 day-old irradiated pupae. On an average, the corresponding controls showed a fertility of 88.75 ± 1.92 .

b) Irradiated female x normal male : When the adult females emerging from 5-7 day old irradiated pupae were allowed to mate with normal males, the fertility of the females was completely inhibited. However, a hatching was 24.47% was realized case eggs laid by females emerging from 7 day old treated pupae. Further, it was noticed that the ovarian development was greatly reduced in the adults resulting from 5 and 6 day old irradiated pupae. The data clearly indicate that a minimum dose of 2 Kr is sufficient enough to inhibit

Table I : Effect of gamma radiation (2 Kr) on the adult emergence (%), fecundity (No.) and fertility (%) of *Exorista bombycis* when the pupae were irradiated.

Age of Pupa (days)	Adult Emergence (%)	Fecundity (No.) (Mean \pm SE)				Fertility (%) (Mean \pm SE)			
		1MxNF	1FxNM	1Mx1F	NMxNF (Control) @	1F xNM	1FxNM	1Mx1F	NMxNF (Control) @
4	3.0 ± 0.24	0.0	0.0	0.0	485.60 ± 56.20	0.0	0.0	0.0	88.75 ± 1.92
5	69.0 ± 2.64	413.33 ± 67.40	0.0	0.0	485.60 ± 56.20	0.0	0.0	0.0	88.75 ± 1.92
6	86.00 ± 1.0	456.30 ± 69.20	0.0	0.0	485.60 ± 56.20	0.0	0.0	0.0	88.75 ± 1.92
7	87.33 ± 19.20	418.66 ± 19.20	29.33 ± 1.52	0.0	485.60 ± 56.20	22.70 ± 2.12	24.47 ± 2.12	0.0	88.75 ± 1.92
SE \pm	1.34	36.53			-				-
CD at 5%	3.80	73.44							

@ : An adult emergence of 90.20 ± 1.40 % was recorded in control; I : Irradiated male (M) or female (F); N : Normal male or female; M : Male; F : Female.

fertility when the adult females emerging from the irradiated pupae of 5 and 6 day old mate with normal males.

Gamma irradiation of pupae induced sterility in males of *A. ludens* with 5 Kr (Rhode *et al.*, 1961), *M. domestica* with 2.85 Kr (Schmidt *et al.*, 1964) and *C. capitata* with 10 Kr (Steiner *et al.*, 1965). However, only partial sterility was accomplished in both the males and females of *C. hominivorax* (Bushland & Hopkins, 1953) with 8 Kr.

Our findings while agreeing with the results of most of the above workers, differ from those recorded by Bushland & Hopkins (1953) who have reported higher tolerance of females of *C. hominivorax* to gamma radiation compared to the males. Nevertheless, in *E. bombycis* it is interesting to note that the radiation dosage (2 Kr) that induced sterility in males also induced sterility in females.

Brood test

Brood test was performed to record the recovery of fertility, if any, in the males emerging from 5 and 6 day-old pupae which were gamma irradiated with 2 Kr. An average 351 (320-382) and 346 (314-378) eggs were laid when untreated females were mated with sterile males emerging from 5 and 6-day old irradiated pupae (Table II). No hatching was recorded in any of the batches of eggs collected up to 10 days when all the males died. This confirms the fact that there is no recovery in fertility in the males emerging from 5 and 6 day-old pupae treated with 2 Kr gamma radiation.

The findings of the brood test in the present study have revealed that a dose of 2 Kr was sufficient enough to induce sterility in the males of *E. bombycis* throughout the life span of flies emerging from 5 and 6 day-old irradiated pupae when allowed for repeated mating with different virgin female flies. Obviously, this is due to an irreversible impairment in the process of spermatogenesis and spermiogenesis.

Kaufmann & Wassermann (1957) reported that sperm production was permanently altered when 5 day-old pupae of screw-worm fly were irradiated with 7.5 Kr of X-rays. Similarly, Reimann (1967) found that no normal sperms were produced in this insect when exposed to gamma rays at spermatid stage with 6.2 Kr.

Mating competitiveness

The data on mating competitiveness are presented in Table III. In the absence of a sterile male, mating between a normal male and a normal female resulted in an average fecundity of 423.33 eggs, 89.01% fertility and 70.06 F1 progeny. After introducing the irradiated male among the normal males and the normal females, these values altered.

In all the combinations (ratios) (1:1:1, 3:1:1, 5:1:1, 7:1:1, 9:1:1 and 10:1:1 of $\text{I}\delta \times \text{N}\text{N}\text{f}$), no significant difference in the fecundity was recorded. But the fertility and F1 progeny (based on adult recovery) were reduced significantly. At higher ratios of 10:1:1 and 9:1:1, all the eggs laid (352.66 and 332.66, respectively) were found non-viable. At the lowest ratio of 1:1:1, a progeny of 18.78% was recorded which was significantly lower than that of control. Similarly, at 3:1:1 and 5:1:1 and 7:1:1 ratios, the progeny production

Table II : Sterility of the male *E. bombycis* emerging from 5 and 6 day-old irradiated (2 Kr) pupae.

Days of brood	Adult emerging from 5 day-old irradiated pupae		Adult emerging from 6 day-old irradiated pupae	
	Fecundity (No.) (Mean \pm SE)	Sterility (%)	Fecundity (No.) (Mean \pm SE)	Sterility (%)
1	374.00 \pm 37.20	100	350.64 \pm 27.50	100
3	320.00 \pm 19.00	100	378.54 \pm 32.65	100
5	373.00 \pm 11.20	100	318.05 \pm 21.00	100
7	347.48 \pm 18.40	100	315.89 \pm 17.20	100
9	382.78 \pm 21.38	100	314.55 \pm 10.66	100
10	368.90 \pm 15.45	100	379.26 \pm 19.33	100
Control	398.20 \pm 19.21	11.0	398.20 \pm 19.21	11.00

Table III : Mating competitiveness of sterile males of *E. bombycis* with normal male and normal female in different ratios.

Ratio (xM : NM : NF)	No. of eggs/ female (avg)	Hatching (%)	Pupation (%)	Adult emergence (%)
0:1:1	423.33	89.01 (70.67)	78.39 (62.42)	70.06 (56.84)
1:1:1	208.00	25.07 (29.99)	22.31 (28.18)	18.78 (25.67)
3:1:1	253.66	15.62 (21.91)	14.20 (22.13)	13.78 (21.79)
5:1:1	278.66	12.47 (19.80)	11.74 (20.04)	11.47 (19.79)
7:1:1	311.20	10.10 (17.40)	3.10 (10.13)	2.94 (9.87)
9:1:1	332.66	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)
10:1:1	352.66	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)
SE +	49.52	3.65	1.20	0.56
CD at 5%	150.23	11.08	3.64	1.70

Figures in parentheses are angularly transformed values; M : Male; F : Female.

Table IV : Mating efficiency of sterile males following the release of flies in 10:1:1 ratio (sterile male: normal male: normal female) in a rearing house.

Day of observation	No. of silkworms exposed	No. of silkworms infested	Total No. of eggs laid	Sterility (%)
1	3750	201	613	100
2	3750	182	552	100
3	3750	169	339	100
4	3750	213	438	100
5	3750	156	296	100
6	3750	76	163	100

declined progressively and the values stood at 13.78, 11.47 and 2.94%, respectively.

To test verify the above findings, an experiment was also conducted in a silkworm rearing house (20' x 15' x 10') by maintaining the ratio between sterile male, normal male and normal female at 10:1:1. The results are summarized in Table IV. Altogether 3750 silkworm larvae of IV and V instars were exposed for a period of 6 days for oviposition. A total of 2401 eggs were laid on 997 silkworms and all the eggs were found non-viable.

In the field release of sterile males of an insect species, certain females mate with normal males until the area is flooded with sterile males. Therefore, determination of a suitable ratio of sterile male to normal male of *E. bombycis* for field release is essential. In the present study, attempts were made to determine the most suitable ratio by conducting cage and rearing house experiments. In both the experiments, a ratio of 7:1:1 (Irradiated male : Normal male : Normal female) yielded 10% hatching of eggs against no hatching in the treatments of 9:1:1 and 10:1:1. Therefore, a ratio 9:1:1 or greater than that would be necessary for complete loss of reproduction in the field populations of uzi fly.

Our findings are in consonance with those of Qureshi (1966) working on the fruit fly, *D. cucurbitae* at a ratio of 9:1:1 and Manikas & Baldwin (1967) using the olive fruit fly, *D. oleae* at a ratio of 8:1:1 who observed the effective reduction of fly population. But Wong *et al.* (1990) required a higher ratio of 45:1:1 for suppression of *C. capitata* population. In the present study, 25.07 % egg viability was recorded at 1:1:1 ratio, while almost the same degree of viability was recorded at 3:1:1 in the Oriental fruit fly (Steiner *et al.*, 1965).

In the classical sterile male release (SMR) technique, only the males are sterilized and released. But, it has also been found that release of both the sexes is not inferior to the release of males alone (Whitten & Tayler, 1970; Mc Innis & Wong, 1990).

The sterilization of *E. bombycis* through gamma radiation is bound to serve as an effective tool for the suppression of its natural populations as both the sexes of this fly pest can be sterilized by exposing 6 days-old pupae to 2 Kr gamma radiation. Keeping in view a flight range of 2.7 Km in *E. bombycis* (Narayanaswamy *et al.*, 1994), uzi fly management programme using release of sterile populations may prove to be the single most effective tool for uzi fly control if adopted in isolated, uzi fly infested areas. However, in continuous uzi fly infested patches, this method can be combined with the available integrated uzi management package comprising spray of an ovicide (uzicide), placement of an adult chemotrap (uzitrap) and release of an indigenous pupal parasitoid (*Nesolynx thymus*).

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