ผลของรังสีแกมมาที่มีต่อมอดยาสูบ, Lasioderma serricorne (F.)

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บทคัดย่อ

ศึกษาผลของรังสีแกมมาที่มีต่อ ไข่ หนอน ดักแด้ และตัวเต็มวัยของมอดยาสูบค่า ${\rm LD}_{50}$ และ ${\rm LD}_{99}$ ของไข่อายุ 7 ± 1 วัน ภายหลังฉายรังสี 3 วันเท่ากับ 327 และ 834 เกรย์ ที่ปริมาณรังสี 300 เกรย์ ไข่สามารถฟักเป็นตัวหนอนแต่ตายในระยะดักแด้ที่ 800 เกรย์ ขึ้นไปไข่ไม่ฟักทั้งหมด ค่า ${\rm LD}_{50}$ และ ${\rm LD}_{99}$ ของหนอนวัยสุดท้ายอายุ 7 ± 1 วัน ภายหลังฉายรังสี 12 วัน เท่ากับ 505 และ 1,547 เกรย์ ตัวหนอนตายมากขึ้นเมื่อปริมาณรังสีเพิ่มขึ้นที่ปริมาณรังสี 1,500 เกรย์ ขึ้นไปตัว หนอนตายทั้งหมด และที่ปริมาณรังสีต่ำกว่า 1,200 เกรย์ ตัวหนอนเข้าดักแด้แต่ไม่ออกเป็นตัว เต็มวัยค่า ${\rm LD}_{50}$ และ ${\rm LD}_{99}$ ของดักแด้อายุ 4 ± 1 วัน ภายหลังฉายรังสี 12 วัน เท่ากับ 548 และ 1,578 เกรย์ ดักแด้ตายมากขึ้นเมื่อปริมาณรังสีเพิ่มขึ้น ดักแด้ไม่ออกเป็นตัวเต็มวัยที่ 1,500 เกรย์ ที่ 300 เกรย์ ดักแด้ออกเป็นตัวเต็มวัยให้ลักษณะพิการและเป็นหมัน ${\rm LD}_{50}$ และ ${\rm LD}_{99}$ ของตัวเต็มวัยอายุ 3 ± 1 วัน ภายหลังฉายรังสี 12 วันเท่ากับ 931 และ 1,746 เกรย์ ตัวเต็มวัยตายมากขึ้นเมื่อปริมาณรังสีเพิ่มขึ้นที่ 300 เกรย์ ตัวเต็มวัยวางไข่แต่ไข่ไม่ฟักปริมาณรังสีต่ำสุด 300 เกรย์ สามารถยับยั้ง การเจริญเติบโตของมอดยาสูบได้ทุกระยะ

Melanizationเกิดขึ้นภายในตัวหนอนไม่ฉายรังสีภายหลังฆ่าโดยการแช่แข็งตัวหนอนบางส่วน แสดงการเกิด melanization ไม่สมบูรณ์โดยลำตัวของตัวหนอนบางส่วนแสดงสีน้ำตาลเข้มจนถึงสี ดำ และส่วนที่เหลือเป็นสีเหลือง-ขาว หรือ เทา-เหลือง ตัวหนอนฉายรังสีที่ปริมาณรังสี 100, 300 และ 500 เกรย์ ไม่เกิด melanization หรือเกิดเพียงเล็กน้อยการเกิด melanization ในตัวหนอน ระยะที่ 1 และ 2 ลดลงเมื่อปริมาณรังสีเพิ่มขึ้นตัวหนอนวัยที่ 4 เกิด melanizationเพิ่มขึ้นเมื่อ ปริมาณรังสีเพิ่มขึ้น

Effects of Gamma Radiation on the Cigarette Beetle,

Lasioderma serricorne (F.)

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ABSTRACT

Effects of gamma radiation on eggs, larvae, pupae and adult stages of the cigarette beetle were studied. The LD_{50} and LD_{99} of gamma radiation to the eggs (7±1 days) were 327 Gy and 834 Gy respectively on the 3^{rd} day after irradiation. At 300 Gy the eggs were able to hatch but died in the pupal stage. None of the eggs hatched when treated with 800 Gy and other doses. The LD_{50} and LD_{99} to the last instar larvae (7±1 days) were 505 Gy and 1,547 Gy on the 12^{th} day after irradiation. The mortality of last instar larvae varied directly with the irradiation dose, no larvae could survive when irradiated at 1,500 Gy and above. At the dose lower than 1,200 Gy, some pupation was observed but no adult emergence occurred. The LD_{50} and LD_{99} of gamma radiation to pupae (4±1 days) were 548 Gy and 1,578 Gy on the 12^{th} day after irradiation. The pupal mortality varied directly with the dose. No adult emergence was obtained after irradiated at 1,500 Gy. At 300 Gy, the adults emerged with deformity and sterility. The LD_{50} and LD_{99} to adults (3±1 days) were 931 and 1,746 Gy respectively on the 12^{th} day after irradiation. The mortality of adults increased when irradiated at higher dose and none of the eggs hatched after treated adults with 300 Gy. The dosage of 300 Gy was high enough to inhibit the development of all stages.

Melanization occurred in untreated young larvae after killing by freezing. Some parts of larval body became dark brown to black while the rest of the body was yellow- white or greyed-yellow. In the treated young larvae, non-melanization to slight melanization occurred at 100, 300 and 500 Gy. The degree of melanization in treated young larvae decreased with the increasing doses. In treated old larvae, the melanization increased with the increasing doses.

Keywords: Gamma radiation, Cigarette Beetle

INTRODUCTION

Cigarette beetle is the most serious pest of high-value commodities and stored products. It has been exposed to a variety of pesticides over the years; these pesticides include residual products such as synergized pyrethrins, dichlorvos and fumigants such as methyl bromide and phosphine. Typically, residual products persist throughout the storage season or grain shipments for control of insect population growth, whereas fumigants are applied once or twice a year for complete and rapid disinfestation of the commodity.

The Environmental Protection Agency of United States has proposed to eliminate the production and importation of methyl bromide by the year 2001 because of its expected ozone depletion potential⁽¹⁾. Under the Montreal Protocol, the production of methyl bromide in advanced countries must be reduced by 50% in 2001 and 70% in 2003 with a complete phase out by January 2005. In developing countries, they will be allowed to reduce the consumption of this chemical by only 20% in 2005 with the complete out by 2015⁽²⁾. The search for new alternatives to replace fumigants has renewed in the use of radiation techniques in stored product protection⁽³⁾. The advantages of irradiation processing include no undesirable residues in foods, no resistance developed by the insects and no significant changes in the physiochemical properties or the nutritive value of the treat products⁽⁴⁾. Irradiation requires very short exposure times and is limited only by the throughout rate. Irradiation is technically effective in quarantine treatment for stored product. At low doses, irradiation does not cause immediate death of pests⁽⁵⁾. Particularly in products irradiated for quarantine purpose, it must be ascertained that living insects will not be able to survive or proliferate in a new location. Therefore, some methods to detect previous exposure of insect pests to ionizing radiation are needed⁽⁶⁾. The objectives of this study are:

- 1. To study the effects of gamma radiation on eggs, larvae, pupae and adults of the cigarette beetle, *Lasioderma serricorne* (F.).
- **2.** To study the visual changes in melanization on the larval body, which could be useful for determine the difference of irradiated larvae from normal larvae of cigarette beetle, *Lasioderma serricorne* (F.).

MATERIALS AND METHODS

Radiation Effects on Eggs, Larvae, Pupae and Adults

The eggs at 7 ± 1 days old of age were irradiated at 0, 200, 400, 600, 800 and 1,000 Gy. The 4th instar larvae (7 ± 1 days), pupae (4 ± 1 days) and adults (3 ± 1 days) were irradiated at 0, 300, 600, 900, 1,200, 1,500 and 2,000 Gy (dose rate of 2.1 Gy/min) by a Co⁶⁰ gamma irradiator (Gamma Cell 220). One treatment consisted of 100 individuals of each stage with 4 replications. Each stage of insects was placed in plastic tube (4.4 cm x 1.7 cm diameter) and exposed to gamma radiation at the center. A gammachrome was used for dosimeter. Treated and untreated insects were held in the same conditions previously described. After irradiation the larvae were reared in petri dishes (1 cm tall x 9 cm diameter) with crushed corn meal. The mortality after irradiation of all stages, survival period for growth stages and sterility dose for young adults were recorded every day for 12 days. The median lethal dosages (LD₅₀) and ninety nine lethal dosages (LD₉₉) of the gamma radiation to all stages were determined by using probit analysis (7) after making corrections for natural mortality by using Abbott's formula (8).

Radiation Effects on Melanization Process in the 1st, 2nd and 4th Instar Larvae

The first, second and fourth instar larvae were treated at dose of 100, 300 and 500 Gy respectively by gamma irradiator (Gamma Cell 220). A hundred insects of single stage were placed in 4.4 cm x 1.7 cm plastic tube and exposed to gamma radiation at the center. A gammachrome was used for dosimeter. Treated and untreated insects were held in the same conditions previously described. Each treatment consisted of 4 replications. After irradiation, the larvae were reared in petri dishes (1 cm tall x 9 cm diameter) with safflower. In the 1st week after irradiation the larvae were cold-killed by kept in freezer ($-20\pm1^{\circ}$ C) for 24 hours. Larvae were then removed from the freezer and placed on a white background for 2-3 hours at room temperature. Melanization was evaluated visually. The observations on melanization process were made by using stereomicroscope (x20). The size of melanized body portion (black colour) was shown by schematic drawings⁽⁹⁾. An index of melanization was calculated as a mean percentage of larval body encompassed by darkening. The colour was compared with R.H.S. Colour Chart (the Royal Horticultural Society London) as follow: A yellow-white 158 (B-D) colour of larvae = 0.0 (0%), A greyed-yellow 162 (B-D) colour of larvae = 0.33 (33%), A greyed-orange 165 (B)

colour of larvae = 0.66 (66%), A black 202 (A), brown 200 (A-D) and greyed-orange 165 (A) colour of larvae = 1.0 (100%). The data were analyzed as mean values and Duncan's multiple range test, 5% level of probability, was used as the criteria of significance.

RESULTS AND DISCUSSION

Effects of Gamma Radiation on Eggs, Larvae, Pupae and Adults

The effects of gamma radiation on the egg hatchability, pupation and adult emergence are shown in Table 3, 4 and 5. Results on egg irradiation at different doses are shown in Table 1. The egg mortality increased with the increasing dose. At 200 Gy, the percentage of egg hatchability decreased from 71.25 to 39.25 and no sign of pupation. None of the egg hatched after treated with 800 Gy (Table 3). No pupation was observed from the irradiated eggs at any other doses. The percentage of hatchability decreased when increased dose, the treated eggs were grey in colour within 4-5 days after irradiation. The LD_{50} and LD_{99} of gamma radiation to 7 ± 1 days eggs observed on the 3^{rd} day after irradiation were 327 Gy and 834 Gy respectively (Fig. 1).

The results on larval irradiation at different doses are shown in Table 2. The mortality of larvae were found to vary directly with the radiation dose, and no larvae could survive when irradiated at 1,500 Gy and above. At the dose lower than 1,200 Gy, some pupation could be observed but no adult emergence occurred because of their abnormality (Table 4). The colour of the dead treated larvae turned in to black. The LD_{50} and LD_{99} of gamma radiation to 7±1 days larvae on the 12th day after irradiation were 505 Gy and 1,547 Gy respectively (Fig 2).

Results on the pupal irradiation are shown in Table 2. The mortality of pupae also varied directly with the dose. No adult emergence was obtained after irradiated at 1,500 Gy (Table 5). Deformity due to excessive amount of fluid in membranous wings occurred in the treated pupae. The resultant edema caused the hind wings extended and were balloonlike in appearance. These were the most common malformations, the others were deformed elytra, variations in coloration and fused segments⁽¹⁰⁾. When the treated pupae died they turned to black as well. The LD₅₀ and LD₉₉ of gamma radiation to 7 ± 1 days old pupae on the 12^{th} day after irradiation were 548 Gy and 1,578 Gy respectively (Fig 3). The mortality of adults increased when irradiated at higher dose (Table 2) and none of the eggs hatched after the adults were treated with 300 Gy. The LD₅₀ and LD₉₉ of gamma radiation to 3 ± 1 days old adults observed on the 12^{th} day after irradiation were 931 Gy and 1,746 Gy respectively (Fig 4). The mortality of untreated adult was as high as in

treated adult because the longevity of adult was 2-6 weeks (11).

This experiment indicates that the egg was the most susceptible stage to radiation and the adult stage was the most tolerant among all stages. This was due to the Bergonie and Tribondeau's theory which noted that tissues appear to be more radiosensitive when the cells was less differentiated, that the cell was greater proliferative capacity and divide more rapidly. The cell cycle was delay after irradiation, that the mitotic index was reduced. The longest delay was seen for cells irradiated in G_2 , the accumulation of cells in G_2 is connected either with a surveillance mechanism blocking the cell with radiation-induced lesion which prevents cell division, for example chromosome lesions or to a slowing down in the synthesis of proteins necessary for mitosis (12). At lower dose the longevity of insects was reduced, delayed molting, reduction of eggs hatch, inhibition of development, reduction of food consumption, sterility (10). Radiosensitivity depending on the type of cell the germinal stem cells of testis (spermatogonia) are the most radiosensitive (12). At higher dose, immediate mortality was obtained as interpretation of the acute effects of radiation (10). The efficacy of irradiation disinfestation treatment could be measured by prevention of adult emergence when only eggs and larvae are present, and sterility when pupae or adults are present.

Table 1 Effect of gamma radiation on 7 ± 1 days old eggs of the cigarette beetle, (*Lasioderma serricorne*) on the 3^{rd} day after irradiation.

Berriedine) on the 5 day ar	er madation.
Dose (Gy)	% Mortality 1 d
0	0
200	44.56
400	72.63
600	83.16
800	100.0
1,000	100.0

¹ mean of 4 replications.

Table 2 Effect of gamma radiation on 7±1 days old larvae, 4±1 days old pupae and 3±1 days old adults of the cigarette beetle, (*Lasioderma serricorne*) on the 12th day after irradiation.

Dose (Gy)	% Mortality 1 1		
•	Larvae ²	Pupae ²	Adults ²
0	0	0	0
300	47.15	42.47	10.26
600	63.92	64.38	20.51
900	83.92	79.45	26.92
1,200	94.94	90.41	91.02
1,500	100.0	100.0	100.0
2,000	100.0	100.0	100.0

¹ mean of 4 replications.

Table 3 Percentage of hatchability, pupation and adult emergence of the cigarette beetle eggs, (*Lasioderma serricorne*) on the 3rd day after irradiation.

eggs, (Lusio	dernia serricorne, on the 5	day and mradianon.	1
Dose (Gy)	Hatchability (%) ¹ □	Pupation (%)	Adult emergence(%) ¹
0	71.25	65.50	61.75
200	39.25	0	0
400	19.50	0	0
600	12.0	0	0
800	0	0	0
1,000	0	0	0

¹ mean of 4 replications, 100 individuals / replication.

Table 4 Percentage of pupation and adult emergence of the cigarette beetle larvae, (*Lasioderma serricorne*) on the 12th day after irradiation.

Dose (Gy)	Pupation (%) ¹	Adult emergence (%) ¹
0	52.25	47.75
300	37.25	0
600	25.0	0
900	9.75	0
1,200	1.25	0
1,500	0	0
2,000	0	0

¹ mean of 4 replications, 100 individuals / replication.

Table 5 Percentage of adult emergence and adult deformity from pupae of the cigarette beetle, (*Lasioderma serricorne*) on the 12th day after irradiation.

occie, (Eustouerma serricorne) on the 12 day after madiation.			
Adult emergence (%)	Adult deformity (%)		
53.75	12.0		
32.75	64.25		
25.25	78.50		
9.50	88.75		
6.25	95.0		
0	0		
0	0		
	Adult emergence (%) ¹ 53.75 32.75 25.25 9.50 6.25		

¹ mean of 4 replications, 100 individuals / replication.

^{2 100} individuals / replication.

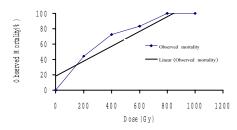


Figure 1 Effect of gamma radiation on 7 days old eggs of the cigarette beetle, (Lasioderma serricorne) observed on the 3^{rd} day after irradiation. Y = 0.0967x + 18.379 R² = 0.877 LD₅₀ = 327 Gy LD₉₉ = 834 Gy

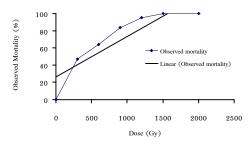


Figure 2 Effect of gamma radiation on 7 days old larvae of the cigarette beetle, (*Lasioderma serricorne*) observed on the 12^{th} day after irradiation. $Y = 0.047x + 26.275R^2 = 0.799$ $LD_{50} = 505$ Gy $LD_{99} = 1,547$ Gy

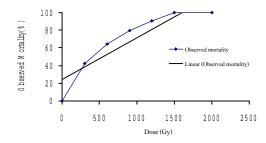


Figure 3 Effect of gamma radiation on 4 days old pupae of the cigarette beetle, (Lasioderma serricorne) observed on the 12^{th} day after irradiation. $Y = 0.0476x + 23.906 R^2 = 0.8283 LD_{50} = 548 Gy LD_{99} = 1,578 Gy$

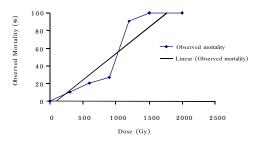


Figure 4 Effect of gamma radiation on 3 days old adults of the cigarette beetle, (*Lasioderma serricorne*) observed on the 12^{th} day after irradiation. Y = 0.0601x - 5.9549 R² = 0.8642 LD₅₀ = 931 Gy LD₉₉ = 1,746 Gy

Effect of Radiation on Melanization Process in the 1st Instar Larvae

The colour of cigarette beetle larvae in control treatment were black, greyedorange, greyed-yellow and yellow-white in ratio of 0.79: 1.83: 1.01: 0.31 respectively, the larvae exhibited greyed-orange and greyed-yellow more than the other colours. Some larvae showed black colour on the body and greyed-yellow on the another half. The mean percent of changing was 59.15 % (Table 6). The larvae treated with dose 100 Gy showed variable colours in ratio of 0.44: 0.67: 2.04: 0.83 from black, greved-orange, greved-yellow and yellow-white colour. Most of the larvae showed greyed-yellow colour more than other colours. Few larvae showed black colour on a part of larval body, while the rest of the body was greyed-yellow. The mean percent was 39.3 % (Table 6). The larvae treated with dose 300 Gy showed the colours in ratio of 0.37: 1.24: 1.56: 0.91 from black, greyed-orange, greyed-yellow and yellow-white colour. The larvae showed greyed-orange and greyedyellow colour more than the other colours. Some larvae showed melanization only partially. The mean percent was 40.88 % (Table 6). The colours of larvae treated at dose 500 Gy are shown in ratio of 0.11: 1,29: 1.46: 1.06 from black, greyed-orange, greyed-yellow and yellow-white colour. Most of the larvae showed greyed-orange and greyed-yellow colour rather than the other colours. Some larvae showed melanization only partially. The mean percent was 36.74 % (Table 6). The degree of melanization in treated larvae was significantly different from untreated larvae, it decreased with the increase in dose.

Effect of Radiation on Melanization Process in the 2nd Instar Larvae

The body colours of the cigarette beetle larvae in control treatment were black, greyed-orange, greyed-yellow and yellow-white in ratio of 0.57: 1.70: 1.61: 0.40. The larvae exhibited greyed-orange and greyed-yellow more than the other colours. Some of larvae were black on one-half of the body and greyed-yellow on another half or showed melanization only partially. The mean percent was 56.58 % (Table 7). The larvae treated with dose 100 Gy showed variable colour in ratio of 0.28: 0.63: 2.33: 0.84 from black, greyed-orange, greyed-yellow and yellow-white colour. Most of the larvae showed greyed-yellow colour more than other colours. Some showed black colour on a part of larval body, while the rest of the body was greyed-yellow. The mean percent was 35.88% (Table 7). At 300 Gy, the treated larvae were black, greyed-orange, greyed-yellow and yellow-white in ratio of 0.14: 1.09: 1.76: and 0.67. The larvae showed greyed-orange and greyed-yellow colour more than other colours. Some larvae showed melanization only partially. The mean percent was 39.45 % (Table 7). The larvae treated with dose 500 Gy showed black, greyed-orange, greyed-yellow and yellow-white colour in ratio of 0.01: 1.27: 2.17: 0.52. Some larvae showed melanization only partially. The

mean percent was 39.40 % (Table 7). The degree of melanization in treated larvae was significantly different from untreated larvae, it decreased with the increasing dose.

Effect of Radiation on Melanization Process in the 4th Instar Larvae

The larvae in control were black, greyed-orange, greyed-yellow and yellow-white in ratio of 0.07: 1.45: 0.85:1.03. The larvae showed greyed-orange and yellow-white more than other colours. Some of larvae were partially darken. The mean percent was 33.36 % (Table 8). The colour ratio of black: greyed-orange: greyed-yellow: yellow-white colour was 0.19: 0.37: 1.39: 0.71 in the larvae treated with dose 300 Gy. The greyed-yellow was the dominant colour. Some larvae showed melanization only partially. The mean percent was 42.66 % (Table 8). The larvae treated with dose 500 Gy showed the colour in ratio of 0.18: 1.89: 0.67: 0.05 from black, greyed-orange, greyed-yellow and yellow-white colour. The larvae showed greyed-orange colour more than other colours. Some of larvae showed melanization only partially. The mean percent was 51.86 % (Table 8). The degree of melanization in treated larvae was significantly different from untreated larvae, it increased when the dose was increased. Delay developing to pupal stage was found in the treated old larvae, this may due to its low metabolic rate. The mean percent of melanization in treated old larvae was higher than in untreated old larvae.

Banasik-Solgala and Stanislaw⁽¹⁴⁾ found the degree of melanization differed significantly between treated and untreated insects. It decreased with increasing dose after treatment in old larvae of the Indian meal moth, *Plodia interpunctella* Hub., the Mediterranean flour moth, *Ephestia (Anagasta) kuehniella* Zell., and the almond moth, *Cadra cautella* Wlk., Ignatowicz and Ibrahaim⁽⁹⁾ reported that when irradiation young larvae of the confused flour beetle, *Tribolium confusum* J. du Val., the melanization was reduced in first week. Great variation of melanization in the untreated old larvae partially obscured the effects of gamma radiation on this process. However, the melanization was considerably reduced in all the experiments involving old larvae. Ignatowicz and Katarzyna⁽¹⁵⁾ reports that after the irradiation treatment with doses ranging from 100 to 500 Gy, the melanization process was significantly inhibited in the 1st and 2nd instar larvae of the Khapra beetle, *Trogoderma granarium* Ev., killed by freezing on the 1st and 2nd week after irradiation. Also irradiation inhibited significantly the melanization of the 4th instar larvae after their death, however the change in melanization of the Khapra beetle larvae cannot be used for indicating previous exposure of these insects to irradiation, because of the great variability in

response of the melanization process to the irradiation treatment.

An important point borne out of this experiment is the need to specify the occurrence of melanization only in the early stage of larval development. This is due to the observation of rather drastic changes in appearance between early and late instars using a given radiation. Because of this, the results on melanization of the larval body, is worth to serve as leases for future work.

Table 6 Effect of radiation on melanization of the body in the 1st instar larvae of the cigarette beetle (*Lasioderma serricorne*), cold-killed in the first week after treatment.

Dose (Gy)	Number of larvae	Mean percent of the body
		encompassed by the melanization
0	400	59.15b ¹
100	400	39.30a
300	400	40.88a
500	400	36.74a

cy = 18.06%

Table 7 Effect of radiation on melanization of the body in the 2nd instar larvae of the cigarette beetle (*Lasioderma serricorne*), cold-killed in the first week after treatment.

beene (Eastouerma serricorne), cold kined in the first week after treatment.		
Number of larvae	Mean percent of the body	
	encompassed by the melanization	
400	56.58a ^{1]}	
400	35.88c	
400	39.45b	
400	39.40b	
	Number of larvae 400 400 400	

cy = 3.38%

Table 8 Effect of radiation on melanization of the body in the 4th instar larvae of the cigarette beetle (*Lasioderma serricorne*), cold-killed in the first week after treatment.

after treatment.		
Dose (Gy)	Number of larvae	Mean percent of the body
		encompassed by the melanization
0	400	33.36a ¹
100	400	49.51b
300	400	42.66ab
500	400	51.86b

cy = 19.03%

¹ Means within a column followed by the same letter are not significantly different (P>0.05), Duncan's multiple range test.

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¹ Means within a column followed by the same letter are not significantly different (P>0.05), Duncan's multiple range test.

CONCLUSION

The cigarette beetles were reared successfully on crushed corn meal in glass jars at 30±2°C and 70±5% RH in the laboratory. Eggs were irradiated at 0, 200, 400, 600, 800 and 1,000 Gy. The percentage of hatchability decreased when increased dose. The eggs hatched after being treated at lowest dose but died in larvae stage. The LD₅₀ and LD₉₉ of gamma radiation to the eggs were 327 Gy and 834 Gy respectively. The last instar larvae were irradiated at 0, 300, 600, 900, 1,200, 1,500 and 2,000 Gy. The mortality of larvae varied directly with the radiation dose. At lower dose, the treated larvae couldn't develop to adult. The LD_{50} and LD_{99} of gamma radiation to the larvae were 505 Gy and 1,547 Gy. The pupae were irradiated at 0,300,600,900,1,200,1,500 and 2,000 Gy. The mortality of pupae varied directly with the radiation dose, At lower dose, the treated pupae delayed molting and the emergence adults were deformed. The LD_{50} and LD_{99} of gamma radiation to the pupae were 548 Gy and 1,578 Gy. The adults were irradiated at 0, 300, 600, 900, 1,200, 1,500 and 2,000 Gy. The mortality of adults increased when irradiated dose was high. The adults that were treated with 300 Gy produced sterilized eggs. The LD₅₀ and LD₉₀ of gamma radiation to adults were 931 Gy and 1,746 Gy. The adult stages were most tolerant to irradiation. The dosage of 300 Gy was high enough for inhibition the development of all stages. At 300 Gy the longevity was reduced, the other results were delayed molting, reduction of egg hatchability and reduction of food consumption and sterility.

The degree of melanization in untreated 1st instar larvae was significantly different from the treated larvae, it decreased with increased dose. The untreated larvae exhibited completely melanization and melanization only partially. Some larvae showed clear lack of melanization. The treated larvae exhibited incompletely melanization and melanization only partially. The mean percent of melanization in control, 100, 300 and 500 Gy were 59.15%, 39.30%, 40.88% and 36.74% respectively. The degree of melanization in untreated 2nd instar larvae was significantly different from treated larvae. In treated larvae, non-melanization to slight melanization occurred at 100, 300 and 500 Gy. The untreated larvae exhibited completely melanization and melanization only partially. Many treated larvae showed greyed yellow colour. The mean percent of melanization in control, 100, 300 and 500 Gy were 56.86%, 35.88%, 39.45% and 39.40% respectively. The degree of melanization in untreated 4th instar larvae was significantly different from treated larvae, it increased with increased dose. Some of untreated larvae showed strong

darkening and other showed only partially. Treated larvae showed many strong darkening and completely greyed-orange and greyed-yellow colours. The mean percent of melanization in control, 100, 300 and 500 Gy were 33.36%, 49.51%, 42.66% and 51.86% respectively. This experiment showed variation of melanization in the untreated and treated old larvae. The treated old larvae prolonged their larval stage while untreated larvae reached their pupal stage.

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