

**BIOLOGICAL EFFECTS OF GAMMA RADIATION ON
STORED PRODUCT INSECTS.**

**4 - RADIATION EFFECTS ON SEX PHEROMONE
PRODUCTION AND PERCEPTION BY THE RUST-RED
FLOUR BEETLE. *TRIBOLIUM CASTANEUM* (HERBST)**

By

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ABSTRACT

Irradiation of the rust-red flour beetle, *T. castaneum* at different doses of gamma radiation considerably affected sex pheromone production by females and perception by males. The production of sex pheromone by virgin females decreased with the increase of radiation doses from 4 to 10 krad., and a dose of 12 krad could almost inhibit pheromone production.

Males were more radiosensitive in their response to sex pheromone; and a radiation dose of 8 krad could brought inhibition of male response.

Irradiating 5 days old pupae severely affected the sex pheromone production and perception by the resulting adult beetles. The effect was very much evident even at the lowest radiation doses (0.5-1 krad). An almost complete inhibition in pheromone production was reached at a radiation dose of 6 krad; but a dose of 3 krad appeared to be enough to prevent male response.

INTRODUCTION

Radiation as a factor affecting pheromone production and perception in insects has attracted the attention of many investigators. Stimann *et al.* (1972) reported that irradiated females of the cabbage looper, *Trichoplusia ni* (Hubne) became increasingly less attractive to males as the radiation doses increased. Almost similar results were obtained by other workers as Fletcher and Glannakakis (1973), on the male fruit fly, *Dacus tryoni* (Froggatt); Hendericks (1974) on the female tobacco bud worm, *Heliothis virescens* (F.) and Earle *et al.* (1978) on the boll weevil, *Anthonomus grandis* (Boheman).

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The female rust-red flour beetle, *Tribolium castaneum* (Herbst) was found to produce a sex pheromone that aroused and attracted males for mating and attracted females for recognition (Hussien and Abdel-Kader, 1984). The present study deals with the effective of gamma-radiation on sex pheromone production and perception that might contribute in the control of this serious stored product insect through the adverse effect on its reproduction.

MATERIALS AND METHODS

A stock culture of *T. castaneum* was maintained in the laboratory according to the technique followed by Hussien and Abdel-Kader (1984). Beetles were sexed as pupae according to Ho (1969); and individuals of each sex were held in separate incubators adjusted at $30 \pm 1^\circ\text{C}$.

Virgin females (4-6 days old) were exposed to gamma radiation doses; 4, 5, 6, 8, 10 and 12 krad. They were extracted by diethyl ether and bioassayed against unirradiated (4-6 days old) male beetles following the technique applied by Burkholder (1970).

Males of the same age were similarly treated and bioassayed against untreated virgin females. Five days old male and female pupae were also irradiated at doses ranging from 0.5 to 6 krad. They were kept for emergence. Adults emerged there-from were bioassayed for sex pheromone production and perception.

A CO^{60} unit of gamma cell Model Nor. 3500 located at the Middle Eastern Regional Radiosotopes center for Arab Countries, Dokki, Giza was used for irradiating insects. The dose rate was 41.45 rad/sec. at the start of experiments. It then decayed to 39.21 rad/sec. at the end of experiments. Correction for radiation source decay was monthly made.

Sex pheromone extraction of females and tests for male response were carried out at 3.00 p.m.

Data obtained were subjected to the confidence intervals for proportions and do the test of quality of two proportions. $P = 0.05$ was taken as the level of probability in assessing significance.

RESULTS

I Effect of irradiating adults

a) On pheromone production by females:

The number of males responded to female extract was taken as a criterion for the amount of pheromone production. Accordingly virgin females irradiated with increasing doses of gamma radiation progressively produced lesser amounts of

Table 1

Male response of *Tribolium castaneum* to sex pheromone extracts of irradiated (4-6 days old) virgin females.

Radiation dose (krad)	Percent of male response			
	Experiment	Control (solvent)	Corrected experiment	Binomial limits
0	80	10	77.78	72.16 — 87.84
4	70	10	66.67	61.02 — 78.98
5	67	14	61.63	57.78 — 76.22
6	55	10	50.00	45.25 — 64.75
8	42	14	32.56	32.33 — 51.67
10	39	14	29.07	29.44 — 48.56
12	18	12	6.82	10.47 — 25.53

pheromone than the non-irradiated females (Table 1). The number of males responded to diethyl ether extract of females irradiated at 4 krad did not differ significantly from that of the control ($Z=1.75$). The level of male response to extract of females irradiated at higher doses (5-10 krad) significantly decreased indicating a lower titer of pheromone production ($Z > 1.96$).

An almost complete inhibition of pheromone production occurred when females were irradiated at 12 krad; the males did not respond to such females extracts.

b) On pheromone perception by males:

The response of irradiated males to sex pheromone extracts of unirradiated virgin female was highly affected by gamma radiation (Table 2).

The level of response of males treated at the lowest radiation dose (4 krad) to sex pheromone extracts of virgin females severely dropped in comparison with that of unirradiated males. The difference was highly significant ($Z>1.96$).

Exposure of males to higher radiation doses (5 and 6 krads) produced significantly more reduction in the level of males response; and an almost completed inhibition of male response was reached at 8 krad.

Radiation Effects on Insects

Table 2

Response of irradiated (4-6 days old) males of *T. castaneum* to sex pheromone extract of virgin females.

Radiation dose (krad)	Percentage of male response			
	Experiment	Control (solvent)	Corrected experiment	Binomial limits
0	80	10	77.78	72.16 — 87.84
4	26	4	22.92	17.40 — 34.60
5	17	4	13.54	9.64 — 24.36
6	12	4	8.33	9.63 — 18.37
8	8	4	4.17	2.68 — 13.32
10	7	4	3.13	2.00 — 12.00

2. Effects of Irradiating Pupae

a) On pheromone production by emerged females:

Comparable with females emerged from untreated pupae, sex pheromone production by females emerged from irradiated pupae was greatly affected by most of the radiation doses tested (Table 3).

Table 3

Response of (4 - 6 days old) males of *T. castaneum* to sex pheromone extracts of virgin females emerged from 5 days old irradiated pupae.

Radiation dose (krad)	Percentage of male response			
	Experiment	Control (solvent)	Corrected experiment	Binomial limits
0	77	12	73.86	68.75 — 85.25
0.5	70	10	66.67	61.02 — 78.98
1	46	10	40.00	36.23 — 55.77
2	45	10	38.89	35.25 — 54.75
3	33	14	22.09	23.78 — 42.22
4	30	10	22.22	21.02 — 38.98
5	27	10	18.89	18.30 — 35.70
6	18	10	8.89	10.45 — 25.53

The lowest radiation dose (0.5 krad) appeared to have a slight effect. This was evident from the high level of male response. Higher radiation doses (1 and 2 krad), on the other hand, severely affected the pheromone production. More increase in the radiation doses (3-5 krad) showed gradual decrease in the percentage of male response to pheromone extracts indicating more reduction in pheromone production. Inhibition of pheromone production appeared to occur when pupae were exposed at a radiation dose of 6 krad.

b) On pheromone perception by emerged males:

The response of males emerged from irradiated pupae to sex pheromone extracts of unirradiated virgin females were also severely affected (Table 4).

Table 4

Response of 4-6 old days males of *T. castaneum* emerged from irradiated 5 days old pupae to sex pheromone extract of virgin females.

Radiation dose (krad)	Percentage of male response			
	Experiment	Control (solvent)	Corrected experiment	Binomial limits
0	77	12	73.86	68.75 — 85.25
0.5	50	10	44.44	40.20 — 59.80
1	27	14	15.12	18.30 — 35.70
2	25	14	12.79	16.51 — 33.39
3	12	10	2.2	5.63 — 18.37
4	10	6	4.26	4.12 — 15.88
5	7	6	1.06	2.00 — 12.00

The level of male response decreased sharply and significantly ($Z > 1.96$) when pupae were exposed at doses of 0.5—2 krad. Exposure of pupae to 3 krad appeared enough to cause inhibition of pheromone perception by the emerged males. Higher doses up to 8 krad gave different but nonsignificant levels of response.

DISCUSSION

It has become evident that in many species of insects pheromones play a significant role in bringing the two sexes together for reproduction (Wood *et al.*, 1970, Jacobson, 1972 and Struble *et al.*, 1974). Thus, any factor adversely affect sex pheromone production on perception by an insect pest may bring about a reduction in its population, simply by affecting its chance to mate and reproduce or by sterilization.

The effect of radiation on the production and perception of sex pheromone may have, as revealed in the present study, an important factor in radiation sterilization studies.

The radiation dose require to reduce or inhibit sex pheromone production *T. castaneum* considerably affects its attractiveness to males. The latters are more radiosensitive concerning their perception to the sex pheromone produced by virgin females.

We do not know whether the changes observed in *T. castaneum* are related to the vulnerability of the nervous system to radiation or radiation damaged pheromone producing glands of the females and loss of chemoreceptive ability of reales. Wharton and Wharton (1975) foud that 1000 rad of radiation of the fast neutrons produced by a 2 meV de Graff accelerator (Model AK) permanently inhibited oothecal formation but only temporarily inhibited pheromone production in females of the American cockroach, *Periplaneta americana*. Menon (1978), on the other hand, indicated that a dose of radiation (14 krad) that was sufficient to induce 99.9 percent sterility in males of the cockroach *Neuphoeta cinera* (Olivier) did not inhibit the pheromone production but did interfere with normal mating behaviour patterns. At higher doses, gamma radiation could have severaly injured the cells synthesizing pheromone, which may account for the decreased pheromone activity.

The present study on *T. castaneum* also indicates that sex pheromone production and preception by beetles emerged from irradiated pupae are more radiosensitive than when beetles themselves are irradiated with similar doses. Again, males are more sensitive than females. Similarly, Fletcher and Glannakakis (1973) studied the effect of sterilizing doses of gamma radiation upon pheromone production in male fruit fly, *Dacus tryoni* (Froggatt). They showed that the exposure of the pharate adult on the ninth day in puparium to 8 krad no detectable effects on pheromone production but the exposure to 5 krad on the fifth day in puparium caused a significant decrease in pheromone production. Hendricks, (1974) showed that females of the tobacco budworn, *Heliothis virescens* (F.), when irradiated as adults with radiation doses ranging from 10-50 krad were at least as attractive as untreated females. Females irradiated at 10 krad as 4 day-old pupae before eclosion had reduced attractiveness; while a considerably greater reduction was reached at 50 krad.

From the foregoing discussion it can be concluded that the effect of gamma radiation on sex pheromone production and perception is an important factor that can be made use of in the control of *T. castaneum*. The effect will be through preventing the two sexes from coming together for mating and reproduction. It is recommended to apply radiation to pupae as they need lower effective doses than adult beetle.

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التأثيرات البيولوجية لأشعة جاما على آفات الحبوب المخزونة
(٤) تأثير الأشعاع على إنتاج واستجابة فيرمون الجنس
في خنفساء الدقيق الصدئية

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كان لتعريض خنفساء الدقيق الصدئية لأشعة جاما تأثيراً ملحوظاً على مقدرة الأناث لافراز فيرمون الجنس ومقدرة الذكور للاستجابة له .

إنخفض معدل إنتاج الفيرمون للأناث التي لم يسبق لها التزاوج مع زيادة معدل الجرعة الأشعاعية من ٤ - ١٠ كراد وكانت الجرعة ١٢ كراد كافية لاثباط إنتاجه . كما كان للأشعاع تأثيراً على مدى استجابة الذكور لفيرمونات الجنس المنتجة من اناثها حيث كانت الاستجابة معدومة تماماً عند المستوى الأشعاعي ٨ كراد . وكان لتعريض عذارى تبلغ من العمر خمس أيام تأثير على الخنافس الناتجة من حيث مقدرتها على الإنتاج أو الاستجابة لهذا الفيرمون حيث كان هناك اثباط كامل لإنتاج الاناث للفيرمون عند جرعة ٦ كراد بينما كانت الجرعة ٢ كراد كافية لاثباط استجابة الذكور للفيرمون تماماً .