Crawford, D. L. 1927. Investigation of Mexican fruit fly (*Anastrepha ludens* Loew) in Mexico. Mon. Bull. California Dep. Agr. 16 (8): 422–45.
Flitters, N. E., P. S. Messenger, and C. N. Husman. 1956. Bioclimatic cabinets used in studies on the Mexican fruit fly and the pink bollworm. USDA, ARS-33-33. 12 p.
Hoidale, P. A. 1952. The Mexican fruit fly. USDA Yearbook of Agr. 1952: 559–62.
Myers, K. 1952. Oviposition and mating behavior of the Queensland fruit fly (*Dacus strumeta*) *tryoni* (Frogg). Australian J. Sci. Res. Ser. B. Biological Sciences 5: 264–81.
Rhode, R. H. 1957. A diet for Mexican fruit flies. J. Econ. Entomol. 50 (2): 215.
Roan, C. C., N. E. Flitters and C. J. Davis. 1954. Light intensity and temperature as factors limiting the mating of the oriental fruit fly. Ann. Entomol. Soc. Amer. 47 (4): 593–4.
Stone, A. 1942. The fruit flies of the genus *Anastrepha*. USDA Misc. Pub. 439. 112 p.

Some Effects of Gamma Radiation and a Chemosterilant on the Mexican Bean Beetle[1]

T. J. HENNEBERRY, F. F. SMITH, and W. L. MCGOVERN
Entomology Research Division, Agr. Res. Serv., USDA, Beltsville, Maryland

ABSTRACT

Gamma radiation or the alkylating agent apholate sterilized male and female Mexican bean beetle, *Epilachna varivestis* Mulsant. With adults the effect was produced with ionizing radiation at 8 or 16 kr. Pupae were more susceptible—complete sterilization of females was obtained at doses of 1, 4, 8, or 16 kr and males were sterilized at doses of 4, 8, or 16 kr. Larvae were more susceptible to direct radiation effects than pupae and pupae more susceptible than adults.

Adult male or female bean beetles dipped in an aqueous solution of 0.5% apholate or confined for 48 hours on bean foliage sprayed with the same concentration in water were completely sterilized. Untreated female beetles mated with treated males (irradiated or exposed to apholate at effective doses) deposited about the same number of eggs as females of untreated pairs, but no eggs hatched. Treated female beetles (irradiated or exposed to apholate at effective doses) mated with untreated males deposited few or no eggs. Adult beetles of either sex irradiated in the pupal or adult stage or apholate-treated adults were shorter lived than untreated beetles. Untreated female beetles mated first with irradiated or apholate-treated males produced sterile eggs, but a subsequent mating with untreated males resulted in the production of viable eggs.

Bushland and Hopkins (1951, 1953) investigated the biological effects of ionizing irradiation on screwworm flies, *Cochliomyia hominovorax* (Coquerel). The successful application of these findings was demonstrated by the control of this insect pest through the release of radiation-sterilized males (Lindquist 1955, Knipling 1955, Baumhover et al. 1955). The possible use of the sterile-male technique for the control of other insect pests has been discussed by Knipling (1959, 1960). Chemical compounds which produced an effect in genetic material similar to radiation have been known since 1898 (Alexander 1960). Recently one of these materials has been shown to be a promising house fly, *Musca domestica* L., sterilant (LaBrecque 1961).

The present paper is a report of laboratory studies conducted in 1961 at Beltsville, Maryland, to determine the effect of ionizing radiation and a chemosterilant on the longevity and fertility of the Mexican bean beetle, *Epilachna varivestis* Mulsant.

METHODS AND MATERIALS.—Bean beetles were collected in the field in the larval stage. Adults, irrespective of treatment, were sexed within 8 hours after emergence.

Gamma Irradiation.—Beetles of both sexes in different stages of development were treated in a 21.8 c. cobalt 60 source of gamma radiation calibrated at about 320 r per minute. Fully fed larvae, 3-day-old pupae, or 2- to 3-day-old adults were exposed to doses of 1, 4, 8, or 16 kr.

Insects irradiated in the adult stage were paired individually with the opposite sex immediately after treatment. The immature stages were treated and held until adults emerged, then paired with untreated individuals of the opposite sex.

Chemosterilant.—Groups of 2- to 3-day-old adult male or female beetles were dipped for 5 min in a 0.5% aqueous solution of apholate, or confined for 48 hours on lima bean foliage which had been treated with apholate (0.5% in water). Beetles dipped in apholate were paired with the opposite sex on the same or following day after treatment. Beetles confined on treated foliage were paired after the 48-hour feeding period.

After treatment by either method, the insect pairs were confined on living bean plants. Fresh plants were supplied every other day. Egg masses on the "old" plants were retained to determine viability of the eggs. If no hatching occurred in 2 weeks, eggs were considered nonviable.

The experiments were concluded after 36 days for irradiated insects and 38 days for the chemically treated group. Each experiment was conducted in a randomized block design of 6 replications and the entire radiation experiment was duplicated.

RESULTS.—*Gamma Radiation.*—Larvae were more sensitive to direct radiation effects than pupae (Table 1). Fully fed larvae exposed to 4, 8, or 16 kr were killed. Larvae exposed to 16 kr died within 14 to 16 hours after treatment. Because of the high mor-

[1] Received November 27, 1962; accepted for publication September 17, 1964. This paper was misplaced during a time of changes in the editorial staff; apologies for delayed publication are herewith tendered.—*Managing Editor.*

Table 1.—Survival of Mexican bean beetles exposed to gamma radiation as larvae or pupae.

Dosage (kr)	No. of larvae tested	Percent survival to adult	No. of pupae tested	Percent survival to adult
Control	20	100	24	92
1	17	88	22	100
4	23	0	22	91
8	20	0	22	68
16	20	0	23	83

tality of treated larvae, further studies were not conducted with this stage of the insect in the present experiments. Some mortality occurred when pupae were irradiated, but males and females were sterilized at a dose (4 kr, Tables 1 and 2) which apparently did not affect emergence.

Untreated females mated with males irradiated at doses of 4, 8, or 16 kr in the pupal stage or doses of 8 or 16 kr in the adult stage deposited about the same number of eggs as females of untreated pairs, but none of the eggs hatched (Table 2). Two exceptions to the results were observed: Untreated females mated with males exposed to 16 kr in the pupal stage deposited fewer eggs than females mated to males exposed to 1 kr; the latter females also deposited more eggs than the females of the untreated pairs. This second result remains unexplained; however, a previous report (Howard and English 1924) indicated extreme inherent variation in oviposition by females of the Mexican bean beetle. These authors also found that the viability of deposited eggs was approximately 46% to 52%. The viability of eggs from control insects in the present study was approximately 29%. This may have resulted from less optimum rearing conditions or sampling from a population of inherent lower egg viability. In the latter case, the radiation-dosage fertility response reported may be greater than

Table 2.—Number and viability of eggs produced by Mexican bean beetles exposed to gamma radiation in pupal or adult stage (means of 12 insect pairs).

Mating combination tested and dosage (kr) for treated sex	Mean number of eggs: Deposited[a]	Mean number of eggs: Hatched
Pupal stage		
Treated males × untreated females		
1	350 d	63
4	337 cd	0
8	222 bcd	0
16	141 ab	0
Control	168 abc	23
Treated females × untreated males		
1	123 ab	0
4	7 a	0
8	55 ab	0
16	0 a	0
Control	365 de	88
Adult stage		
Treated males × untreated females		
1	374 de	96
4	542 e	23
8	546 e	0
16	367 de	0
Control	398 de	95
Treated females × untreated males		
1	366 de	67
4	37 a	37
8	0 a	0
16	21 a	0
Control	326 cd	50

[a] Means not followed by the same letter or common letters are significantly different, Duncan's multiple range test.

Table 3.—Longevity of Mexican bean beetles exposed to gamma radiation in pupal or adult stage (means of 12 insect pairs).

Dosage (kr)	Mean age[a] in days at death: Males	Mean age[a] in days at death: Females
Pupal stage		
1	26 de	28 c
4	18 bcd	23 abc
8	8 a	21 abc
16	9 ab	9 a
Control	36 f	36 c
Adult stage		
1	36 f	28 c
4	24 cd	19 ab
8	15 abc	29 c
16	16 bcd	25 bc
Control	34 ef	31 c

[a] Means not followed by the same letter or common letters are significantly different, Duncan's multiple range test.

Table 4.—Fertility of adult Mexican bean beetles dipped in aqueous solution of 0.5% apholate or confined for 48 hours on lima bean foliage treated with an aqueous spray of 0.5% apholate (means of 12 insect pairs).

Mating combination	Mean number of eggs: Deposited[a]	Mean number of eggs: Hatched
Dip		
Treated males × untreated females	411 a	0 b
Treated females × untreated males	50 b	0 b
Foliage spray		
Treated males × untreated females	334 a	0 b
Treated females × untreated males	13 b	0 b
Control	387 a	220 a

[a] Means not followed by the same letter or a common letter are significantly different, Duncan's multiple range test.

Table 5.—Longevity of Mexican bean beetle adults dipped in aqueous solution of 0.5% apholate or confined on foliage sprayed with 0.5% of apholate (means of 12 insect pairs).

Treatment	Mean age[a] in days at death: Males	Mean age[a] in days at death: Females
Dipping	21 a	28 a
Feeding on foliage	11 b	7 b
Control	32 a	36 a

[a] Means not followed by the same letter or a common letter are significantly different, Duncan's multiple range test.

would be expected from insects of an inherently higher egg viability population.

Females exposed to 1, 4, 8, or 16 kr in the pupal stage and mated with untreated males deposited only a few eggs, or no eggs when exposed to the highest dose (Table 2). The eggs produced by beetles exposed to all the lower doses were sterile.

Females exposed to 8 kr in the adult stage and mated with untreated males deposited no eggs; when exposed to 16 kr, only one female deposited a few eggs, none of which hatched.

High mortality of male and female beetles was observed after exposure to gamma radiation in the adult or pupal stage (Table 3). Males treated in the pupal or adult stage were affected at doses of 4, 8, or 16 kr. Males were also shorter lived when exposed in the pupal stage to 1 kr. The results with females treated in the adult stage were erratic. Adult female beetles exposed to 4 kr were shorter lived than untreated beetles, but those exposed to higher doses (8 or 16 kr) were not affected. Females exposed to 16 kr in the pupal stages were shorter lived than untreated females or females exposed to 1 kr in the pupal stage.

Chemosterilant.—Untreated females mated with apholate-treated males (dipped or fed on treated foliage) produced large numbers of eggs, none of which hatched (Table 4). Apholate-treated females (dipped or fed on treated foliage), and mated with untreated males produced only a few eggs, none of which hatched.

Males or females that fed on treated foliage were shorter lived than untreated beetles (Table 5). Beetles dipped in 0.5% apholate were also shorter lived than untreated insects, but the results were variable and the differences not significant.

Multiple Mating.—Untreated female beetles mated with treated males (radiation or apholate) deposited sterile eggs for about 5 weeks. A subsequent mating with untreated males resulted in the production of viable eggs (Table 6).

Table 6.—Viability of Mexican bean beetle eggs from untreated females mated first with irradiated or apholate-treated males and later with untreated males (means of 10 insect pairs).

Treatment of males used in first mating	After first mating with treated male: No. eggs laid in 5 weeks	After first mating with treated male: No. viable eggs	After second mating with untreated male: No. eggs laid in 2 weeks	After second mating with untreated male: No. viable eggs
Irradiated at 4 kr	623	0	33	33
Dipped in 0.5% apholate	815	0	293	242

REFERENCES CITED

Alexander, P. 1960. Radiation-imitating chemicals. Sci. Amer. 202 (1): 99–108.

Baumhover, A. H., A. S. Graham, B. A. Bitter, D. E. Hopkins, W. D. New, F. H. Dudley, and **R. C. Bushland.** 1955. Screw-worm control through release of sterilized flies. J. Econ. Entomol. 48: 462–6.

Bushland, R. C., and **D. E. Hopkins.** 1951. Experiments with screw-worm flies sterilized by X-rays. J. Econ. Entomol. 44: 725–31.

1953. Sterilization of screw-worm flies with X-rays and gamma rays. J. Econ. Entomol. 46: 648–56.

Howard, N. F., and **L. L. English.** 1924. Studies of the Mexican bean beetle in the southeast. USDA Bull. 1243, 5 p.

Knipling, E. F. 1955. Possibilities of insect control or eradication through the use of sexually sterile males. J. Econ. Entomol. 48: 459–62.

1959. Sterile-male method of population control. Science 130: 902–5.

1960. Use of insects for their own destruction. J. Econ. Entomol. 53: 415–20.

LaBrecque, G. C. 1961. Studies with three alkylating agents as house fly sterilants. J. Econ. Entomol. 54: 684–9.

Lindquist, A. W. 1955. The use of gamma radiation for control or eradication of the screw-worm. J. Econ. Entomol. 48: 467–9.

Effect of pH on Sterilizing Activity of Tepa and Metepa in Male House Flies[1, 2]

A. B. BORKOVEC, SHEN CHIN CHANG, and A. M. LIMBURG

Entomology Research Division, Agr. Res. Serv., USDA, Beltsville, Maryland

ABSTRACT

The sterilizing activity of tepa and metepa solutions (buffered to pH 6–8) injected into male house flies, *Musca domestica* L., was determined. No significant variation of activity with pH of the solution was encountered. The degradation of aqueous solutions of tepa and metepa adjusted to pH 4 with sulfuric acid was followed by thin-layer chromatography and bioassay. The results indicated a similarity in the degradative pathway of the two chemosterilants. It was concluded that in partially degraded solutions the sterilizing activity was proportional to the contents of intact tepa or metepa rather than to the total contents of aziridine function.

It has been reported (Beroza and Borkovec 1964) that some of the highly active insect chemosterilants containing aziridinyl groups were very sensitive to acidic media and that their solutions decomposed rapidly at low pH values. An important degradation product of acidified tepa solution is ethylenimine, which was shown to be inactive as a chemosterilant when injected into male house flies, *Musca domestica* L. It appeared then, that the decomposition of tepa to ethylenimine decreased the sterilizing effect and

[1] Received November 20, 1963; accepted for publication April 13, 1964.

[2] Mention of this proprietary product does not necessarily imply its endorsement by the U.S. Department of Agriculture.