

FIG. 2.—Electroantennogram responses of male and female southern pine beetles to the Control (C), 1, 10, and 100  $\mu\text{g}$  of female pheromone (frontalin) per 10  $\mu\text{l}$  of 70% ethanol, and undiluted frontalin (ca. 10,000  $\mu\text{g}$ ). Mean responses of 5  $\delta$  (●—●) and 5  $\text{f}$  (○—○) beetles,  $\pm$  SE.

shows that EAG amplitude increased with an increasing stimulus concentration. These data are similar to results obtained from lepidopterous insect species in response to increasing concentrations of odor stimuli (Boeckh et al. 1965, Payne et al. 1970, Schneider 1969).

The results from this study support the conclusions obtained from field experiments that both male and female beetles are responsive to frontalin (Renwick and Vité 1968). The fact that frontalin elicits electrophysiological and behavioral response from both male and female beetles provides support to the belief that frontalin is an aggregation pheromone (Vité and Renwick 1968), and not a sex

pheromone. In general sex pheromones are released by one sex only and induce a response in the other sex only (Shorey et al. 1968). Aggregation pheromones, however, are often only released by one sex of a species but they cause both sexes to move toward the source (Shorey et al. 1968).

#### ACKNOWLEDGMENTS

I thank Dr. J. P. Vité, D. L. Williamson, and other members of the Boyce Thompson Institute of Plant Research for their assistance and for providing the compounds used in this work, and Dr. D. Schneider of the Max-Planck Institute für Verhaltensphysiologie for review of the manuscript and his helpful comments.

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## Effect of Post-Treatment Temperature upon Worker Honey Bees Exposed to Varying Doses of $^{60}\text{Co}$ Gamma Radiation<sup>1</sup>

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#### ABSTRACT

The mean survival time of non-irradiated adult worker honey bees, and bees exposed to 3700 or 5000 rad of  $^{60}\text{Co}$  gamma radiation is inversely proportional to the

post-irradiation temperature. Radiation reduced the life-span by 50% at the lower temperature (16°C) and up to 75% at higher temperatures (25° and 33°C).

As early as 1930 it was noted that the duration of survival of an irradiated cell was inversely propor-

tional to its activity after irradiation (Bacq and Alexander 1961). Since that time numerous studies have been made on factors that modify radiation response at the cellular and organismic level, and have included research with various insects. A complex assortment of pre-irradiation, during-irradiation and

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post-irradiation conditions when uncontrolled can seriously alter the outcome of experiments, and make it difficult to compare data from one experiment to another. Factors such as age, strain, stage of development, crowding of individuals, ambient gas composition, temperature, rate of metabolism, irradiation dose rate, kind of radiation, and many others have been examined (International Atomic Energy Agency 1963, 1965, 1967, 1969). In the present paper we report the results of experiments to determine the effect of subjecting irradiated adult worker honeybees at low, intermediate, and high temperatures after irradiation.

#### MATERIALS AND METHODS

**Insects.**—Adult worker honeybees of Italian stock supplied by the USDA Bee Breeding Investigations Laboratory, Baton Rouge, La., were used for the 2 series of tests described in this paper. The first series was started Jan. 17, and the second Dec. 3, 1968. Worker bees were put into cages  $4 \times 4.5 \times 1.5$  in.; sides of 8-mesh hardware cloth, and ends, tops and bottoms of wood. An opening in the top held an inverted 20-ml feeder vial and an opening was made in the bottom for removing dead bees. Food consisted of a boiled mixture of 2 parts sugar to 1 part water. Approximately 25 g of bees were placed in each cage in the 1st test series and 20 g in the 2nd series. Three irradiated and 3 control cages were prepared for each temperature in the 2 series of tests.

**Irradiation Techniques.**—All irradiations were made with a 5000-Ci water-shielded  $^{60}\text{Co}$  irradiator. The  $^{60}\text{Co}$  source consisted of a right cylinder of 32 cobalt rods into which a closed metal container was lowered. The dose rate at the time of these experiments was 1800 rad/min, as determined by Fricke dosimetry (Swallow 1960). An aeration line forced fresh air through the chamber to decrease the ozone content

which formed during irradiation (Kertesz and Parsons 1963).

Cages of bees to be irradiated were placed in the container and lowered through the water into the source for predetermined intervals. To each cage was attached a 15-ml glass vial containing Fricke dosimetry solution. The change in optical density of the solution after irradiation was measured with a Beckman-DU spectrophotometer, and appropriate calculations of total absorbed dose (in rads) were then made. Dose in rads is given in Table 1.

**Posttreatment conditions.**—The irradiated and non-irradiated control cages of bees were held at each of 3 temperatures in each series of tests:  $33 \pm 0.5$ ,  $25 \pm 0.5$ , and  $16 \pm 0.5^\circ\text{C}$ . Daily observations were made until all bees were dead, except for the control cages in the 2nd series at  $25^\circ\text{C}$  when the observations were discontinued after 66 days. Dead bees were removed and counted, and sugar syrup was added to the feeder when necessary.

**Analysis of data.**—The  $LT_{50}$  values with 95% confidence limits were obtained using the probit analysis computer program developed by Daum (1970). Where dosage-mortality regressions were non significant, estimated  $LT_{50}$  values were used because their values were in close agreement with corresponding calculated data.

#### RESULTS AND DISCUSSION

The mortality curves (cumulative death rate) of irradiated and control bees in series 1 held at the 3 post-irradiation temperatures are shown in Fig. 1. Survival of the control and irradiated bees was inversely proportional to the post-irradiation temperature. For any 1 temperature, the mortality of irradiated bees was more rapid than the non-irradiated controls. Radiation reduced the lifespan by 64–68% at  $33$  and  $25^\circ\text{C}$  and by 50% at  $16^\circ\text{C}$  when compared with

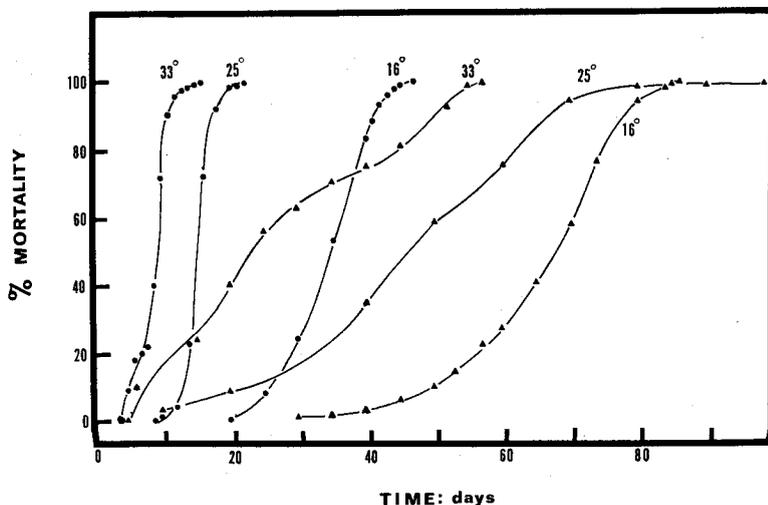


FIG. 1.—Cumulative death rate of irradiated and control honey bees at different temperatures. Dot = irradiated, triangle = controls.

Table 1.— $LT_{50}$  values (in days) with 95% confidence limits for irradiated and control worker honey bees at different posttreatment temperatures.<sup>a</sup>

Test series	33°C				25°C				16°C			
	Dose (rads)	$LT_{50}$	Upper	Lower	Dose (rads)	$LT_{50}$	Upper	Lower	Dose (rads)	$LT_{50}$	Upper	Lower
1	0	21.9	23.6	20.2	0	48.0 <sup>b</sup>			0	67.5 <sup>b</sup>		
	4959	9.3			4960	15.2 <sup>b</sup>			4538	33.6	36.4	30.9
2	0	26.9	43.1	13.4	0	92.1	460.1	61.3	0	29.6	30.4	28.8
	3783	10.8	13.1	8.4	3764	16.2	21.7	4.7	3688	25.2 <sup>b</sup>		

<sup>a</sup> All figures are average of 3 replicates for dose and  $LT_{50}$ .

<sup>b</sup> Estimated value—a nonsignificant regression was obtained in probit analysis.

controls at those same temperatures. Essentially the same results were obtained in the series 2 experiment except that the control bees at 16°C inexplicably died more rapidly than controls at 25°C. Mortality curves for the irradiated bees at all 3 temperatures were very similar for both series 1 and 2.

The mean survival time ( $LT_{50}$ ) for irradiated and control bees at the different temperatures was calculated, and these data are summarized in Table 1. Although the radiation doses of the 2 series differed by approximately 1200 rad, the  $LT_{50}$  for bees held at 33°C and 25°C at both dosages was quite similar. Bees held at 16°C in series 2 had a shorter  $LT_{50}$  at the lower dose. This inconsistency in survival time of this group also was observed in the control bees held at 16°C in series 2 as noted above. With these exceptions, it appears that within the dose range used, mean survival time is independent of total absorbed dose, and is more a function of ambient temperature.

Surprisingly little research has been done with irradiated honeybees. Goolsby (1968) reported that at 94°F (33.9°C) the length of life of bees exposed to 5000 rad of <sup>60</sup>Co was lowered, and that a reduction in post-irradiation temperature increased the lifespan. However, no quantitative data were given to support these observations in detail. It was further noted by Goolsby (1968) that hives irradiated at 5000 rad underwent a 29% decrease in lifespan in the laboratory, and the hive ceased functioning as a social unit within 21 days under field conditions. Pelerents (1963) also noted shortening of lifespan of workers exposed to 4000, 6000, and 8000 rad of <sup>60</sup>Co gamma radiation. He found no dependence in survival time upon dose rate within this range. All of Pelerents' experiments were conducted at hive temperature (35°C) to simulate natural conditions as closely as possible. The irradiated bees lived an average of 9.25 days compared

with 26 days for controls, and these figures coincide almost exactly with our data in Table 1 for 33°C. No other quantitative data are available to our knowledge for bees maintained at lower temperatures after irradiation.

#### ACKNOWLEDGMENTS

We are grateful to Dr. Norbert M. Kauffeld and Mr. Ross A. Nielson, USDA Bee Breeding Investigations, to Dr. L. D. Newsom, Dr. J. B. Graves, and Mr. Gene Strother, Department of Entomology, Louisiana State University for their assistance, and to the Computer Center staff, Louisiana State University, for probit analysis.

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