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Effects of Radiofrequency Electrical Treatment on Fecundity of *Tenebrio molitor* L. (Coleoptera: Tenebrionidae)[1]

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ABSTRACT

When larvae and adults of *Tenebrio molitor* L. were exposed at sublethal levels to 39-MHz radiofrequency (RF) electric fields, the RF treatment of last-stage larvae and adults reduced female fecundity. The reduction in fecundity was more pronounced with longer periods of exposure and higher electrode voltages. The effect of RF energy on fecundity was more dependent upon treatment of males than upon treatment of females. Exposure of larval stages to RF energy resulted in a reduction in both the size of the ovaries and the number of eggs which developed in females that emerged from treated larvae.

The influence of radiofrequency (RF) energy on reproduction in insects has received little attention. Reductions in the number of progeny produced by RF-treated adults have been reported for *Sitophilus oryzae* (L.) and *Tribolium castaneum* duVal (Whitney et al. 1961) and for *S. granarius* (L.) (Thiem and Hank 1966). The present study was conducted to determine the effects of RF treatment on the reproductive potential of male and female *Tenebrio molitor* (L.) after exposure of larvae and adults to RF electric fields.

MATERIALS AND METHODS

Cultures of *T. molitor* were maintained in battery jars in a cabinet with temperature and RH maintained at 30±3°C and 70–75%, respectively. The larvae and adults were confined in polystyrene holders and exposed at sublethal levels to continuous-wave 39-MHz RF electric fields using selected electrode voltages and exposure times. The equipment and the polystyrene holders used were the same as those described by Rai et al. (1971). Adults were exposed in the same type of holders used earlier for pupae (Rai et al. 1971).

For larval treatment studies, 3 groups of 200 larvae that were 30, 60, or 90 days old were exposed to RF electric fields at an electrode voltage of 1.5 kV for 12 s and maintained in petri dishes until the adults emerged. The emerging adults were sexed immediately and isolated before pairing. Then 15 pairs from each age group were set up in individual petri dishes, and the number of eggs laid and the percentage of hatch were recorded for 7 wk. Also, 25 additional females from each group were dissected 10 days after emergence, and the number of chorionated eggs in the egg calyces was counted under the dissecting microscope.

To determine the effects of RF treatment on the reproductive potential of *T. molitor,* 15 pairs of each of 4 mating combinations were maintained in individual petri dishes. The combinations were as follows: 1) treated male × treated female; 2) treated male × untreated female; 3) untreated male × treated female; and 4) untreated male × untreated female. The total number of eggs laid and the percentage of hatch were recorded during a 5-wk period. Also, larvae from eggs deposited by females of the first mating combination were held in containers until the adults emerged. Mean egg production and percentage of hatch were then recorded for 20 pairs of these adults during a 6-wk period. In addition, 25 adults exposed to RF energy were dissected 3 days after emergence and studied, as were another 25 adults developing from untreated larvae.

RESULTS

The data presented in Table 1 show that the mean number of eggs laid by females in mating combinations that included a treated male (combinations 1 and 2) was significantly less than the number laid by females exposed to RF energy (combinations 1 and 3). Also, mean numbers of eggs oviposited decreased considerably with increasing time of exposure. However, none of the percentages of egg hatch from any of the mating combinations was significantly different from that for the check.

The mean number of eggs laid by the progeny of the RF-treated adults was 142.9, ca. ½ the number oviposited by the progeny of untreated adults (296.9), and was thus significantly different ($P < 0.05$). However, the percentages of egg hatch were essentially the same, 71.0 and 77.7%, for progeny from treated and untreated adults, respectively. Likewise, the mean numbers of eggs laid by adults emerging from treated larvae (81.1–98.1 for the 3 ages) and from untreated larvae (154.4) were significantly different ($P < 0.05$), thus indicating that the RF treatment of *T. molitor* larvae reduced the fecundity of resulting adults. However, there was no significant difference between the percentages of egg hatch for treated (65.8–77.3%) and untreated (82.9%) larvae.

The mean number of chorionated eggs in adult females treated at the highest RF energy level was lower than the number in untreated females (Table 2). The mean number of eggs observed in the ovaries of adults that emerged from RF-treated larvae of the 3 age groups was essentially the same.

The effect of RF treatment on ovarian development

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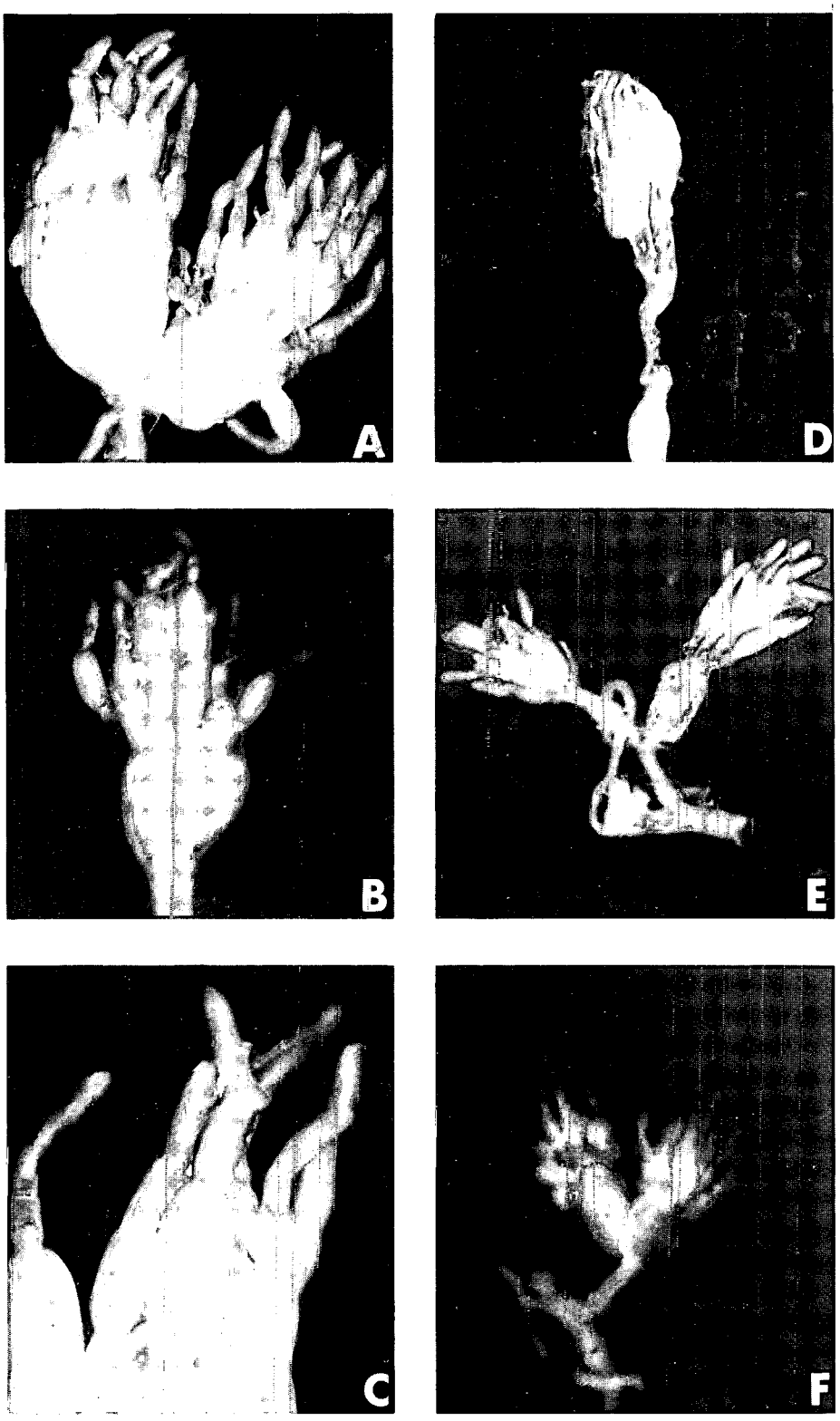

FIG. 1.—Morphological changes in the ovaries of adult *T. molitor* females when larvae of various ages were treated with RF energy at 39 MHz at an electrode voltage of 1.5 kV for 12 s. A.—Ovaries from a normal ♀ 10 days after emergence. B.—A single ovary from a normal ♀. C.—Normal ovarioles. D.—Ovaries from an adult developed from a larva treated at 30 days of age. E.—Ovaries from an adult developed from a larva treated at 60 days of age. F.—Ovaries from an adult developed from a larva treated at 90 days of age.

Table 1.—Mean numbers of eggs laid per *T. molitor* female when mated as indicated and percentage of egg hatch during a 5-wk period after RF treatment of adults at 39 MHz at an electrode voltage of 1.5 kV for indicated exposure times.

	Mating combination[a]							
	Treated ♂ × treated ♀		Treated ♂ × untreated ♀		Untreated ♂ × treated ♀		Untreated ♂ × untreated ♀	
Exposure time (s)	No. eggs	% hatch	No. eggs	% hatch	No. eggs	% hatch	No. eggs	% hatch
1	35.1	84	43.2	81	78.5	85		
2	14.9	73	25.2	75	42.0	73	156.7	84
4	6.5	67	21.2	68	26.5	71		

[a] Within each exposure, differences between mean numbers of eggs laid for mating combinations were all significantly different at the 5% probability level according to the Neuman-Keuls test.

was the same whether the insects were treated in the larval or adult stage. No reduction in the number of ovarioles was observed as a result of treatment, but the size of the ovary was considerably reduced, and the calyx contained few or no eggs. Few developing oocytes were observed in the vitellaria of treated females (Fig. 1 d, e, and f).

DISCUSSION

In this study, the effects of RF treatment were measured indirectly, i.e., by the fecundity of the females in various mating combinations. Hein (1920) reported that *T. molitor* males copulate more than once and that virgin females occasionally deposit eggs, though such eggs do not hatch. Hence, copulation and transfer of viable spermatozoa are essential for normal oviposition in *T. molitor*.

The results indicate that RF treatment affects the fertilization process when either of the sexes is exposed to RF electric fields. Both sexes recovered from the treatment when they were exposed to lower RF-treatment levels; however, adults exposed to high-energy levels rested on their dorsal surfaces after treatment and only gradually recovered or remained in the same position until death. Because females exposed to high-RF-energy levels rested in an inverse position following exposure, copulation was difficult. Also, when males were exposed to high-energy RF-treatment levels, they were physically unable to fertilize the females.

The reduction in fecundity of RF-treated females or of females mated to RF-treated males could therefore result from the physical inability of the males to mate effectively and transfer spermatozoa, from a reduction in the number of developing oocytes in the treated female, or from a reduction in the number and/or viability of spermatozoa transferred by the treated male.

An effect of RF treatment on reproduction in *T. molitor* also manifested itself in the F_1 generation. Probably, treatment lowered the vitality of the treated insects and resulted in physiologically weak progeny.

Oocyte resorption was not found when *T. molitor* females were held as isolated pairs or under crowded conditions (Mordue 1965). Therefore, the observed reduction in the number of developing oocytes was probably caused by the RF treatment.

Table 2.—Mean number of chorionated eggs observed in the egg calyces of *T. molitor* females 10 days post-emergence when adults or larvae were treated with RF energy at 39 MHz.

Stage treated	Age at treatment (days)	Electrode voltage (kV)	Exposure time (s)	No. eggs/female[a]
Adult	3	1.5	4.0	10 a
Adult	3	2.5	4.0	8 a
Adult	3	3.5	4.0	6 b
Adult (untreated)				16 a
Larvae	30	1.5	12.0	13 a
Larvae	60	1.5	12.0	13 a
Larvae	90	1.5	12.0	11 a

[a] Means followed by the same letter were not significantly different at the 5% probability level according to the Neuman-Keuls test.

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