

Testes in *Enchytraeus fragmentosus* Bell

Abstract. *Testes have been localized in the seventh or eighth segment of Enchytraeus fragmentosus* Bell. This species was previously described as lacking gonads.

Bell (1) described *Enchytraeus fragmentosus* as a microannelid lacking reproductive organs and reproducing solely by fragmentation.

Worms received from Bell were cultured at 25°C in petri dishes lined with moist filter paper. Rolled oats was added to the culture dishes as needed. In our laboratory, whole mounts and serial cross sections stained by the Feulgen technique revealed the presence of a pair of testes in the seventh or eighth segment. In squashed preparations, morphologically normal spermatozoa were present in small numbers.

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Reference and Notes

1. A. W. Bell, *Science* **129**, 1278 (1959).
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Endrin Resistance in the Pine Mouse

Abstract. *Wild pine mice with a history of treatment with endrin exhibited a 12-fold greater tolerance to the pesticide than did mice having no history of endrin treatment. A genetic basis for the resistance would be expected, in view of the fact that the resistant mice had been exposed to endrin for 11 years. However, sublethal dosages conferred a degree of resistance on both mouse populations; this indicates the possibility of an inducible resistance.*

Since 1954, endrin (1,2,3,4,10,10-hexachloro-6,7,-epoxy-1,4,4a,5,6,7,8,8a-octahydro-1,4-endo-endo-5,8-dimethanonaphthalene) has been widely used as an effective control measure of the pine mouse *Pitymys pinetorum* in apple orchards (1). However, a number of orchardists have noted that in certain instances endrin lacked effectiveness. We studied the resistance of the pine mouse to endrin. This was assessed by comparing the LD₅₀ values (lethal

dosage for 50 percent mortality) of endrin for susceptible and resistant strains of mice. These values (Table 1) were calculated from regression lines of dosage mortality determined by analysis of probability units. Resistant females and males were 11 and 13 times more resistant, respectively, than their susceptible counterparts were. The pooled LD₅₀ value clearly shows the presence of a 12-fold greater tolerance of endrin by the resistant strain.

The susceptible strain of pine mice was livetrapped during the second week in November 1966 in an orchard located approximately 8 km from Hagerstown, Maryland. With the exception of an application of 0.45 kg of endrin per 0.4 hectare in 1956, this orchard has no previous record of endrin treatment. The resistant strain of mice was captured during the last week in October 1966 4 miles from Berryville, Virginia, in an orchard that has been treated with endrin yearly since 1956. The rate of application was 0.91 kg per 0.4 hectare from 1956 through 1962 and 1.36 kg per 0.4 hectare thereafter. Endrin was applied in 1966 after the test mice were captured. All mice were separated by sex and placed in individual cages with wire bottoms in a temperature-controlled laboratory. A commercial diet and fresh apples were available to the mice at all times. Prior to treatment, the animals were allowed to adapt to laboratory conditions for 7 to 8 days. Juveniles and pregnant females were excluded from the tests.

Endrin (2), dissolved in corn oil, was administered orally on the basis of 1 mg of endrin per 1 kg of body weight. All doses were less than 0.2 ml. Twenty mice were randomly selected for each of five treatments per sex. The amount of treatment for the susceptible mice ranged from 1 to 9 mg of endrin per kilogram of body weight for males and from 2 to 10 mg of endrin per kilogram of body weight for females. For resistant mice the range was 10 to 50 mg of endrin per kilogram of body weight for females and 15 to 55 mg for males. Thus, ten treatments were used for each strain.

We believe that this is the first instance of the development of resistance to a pesticide by mammals in their natural habitat and is in contrast to a study by Ferguson *et al.* (3) who were unable to detect any significant DDT resistance in cotton rats (*Sigmodon hispidus hispidus*) livetrapped from areas treated with DDT. However, Ozburn

Table 1. The LD₅₀ values (in milligrams of endrin per kilogram of body weight) of endrin for susceptible and resistant pine mice. The value for each sex represents 100 mice and five levels of treatment with endrin administered orally in corn oil.

Sex	LD ₅₀ (mg/kg)	
	Susceptible	Resistant
Female	2.93 (2.16-3.97)*	33.28 (28.65-38.66)*
Male	2.98 (1.73-5.12)	39.45 (32.61-47.69)
Pooled	2.97 (2.17-4.08)	36.12 (33.00-39.53)

* Confidence limits 95 percent.

and Morrison (4) were able to produce a twofold tolerance to DDT in a strain of laboratory mice, *Mus musculus domesticus*, by selective breeding of survivors of DDT injections for nine generations. This selected strain also exhibited a cross-tolerance to lindane and dieldrin (5).

If we assume that pine mice are typical of other wild populations with respect to genetic variability, then with direct endrin pressure the basis of the observed resistance could be genetic. Mice with a genetically controlled mechanism to detoxify endrin would thus survive and perpetuate. However, possibly the resistance is not a genetic manifestation but rather an acquired mechanism such as the induction of detoxifying enzymes. To determine if resistance could be induced, we subjected female mice, which had been treated 8 days previously with sublethal doses of endrin, to the calculated LD₅₀ values. None of 18 susceptible females and only 6 of 49 resistant females died. These results suggest the development in the mice of a degree of acquired resistance. It is important to investigate the mechanism of the inducible resistance and the possibility of a genetic aspect of the resistance.

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References and Notes

1. F. Horsfall, Jr., *Virginia Agr. Exp. Sta. Tech. Bull. No. 465* (1953).
2. Endrin was a gift from the Velsicol Chemical Company.
3. D. E. Ferguson, R. L. Callahan, W. D. Cotton, *J. Miss. Acad. Sci.* **11**, 229 (1965).
4. G. W. Ozburn and F. O. Morrison, *Nature* **196**, 1009 (1962).
5. ———, *Can. J. Zool.* **42**, 519 (1964).
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