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RESULTS OF A BIOASSAY TECHNIQUE FOR
GROUND-SPRAYED RODENTICIDES

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ABSTRACT: A laboratory test of toxicity to voles of toxicants sprayed on a soil substrate was used to test brodifacoum, chlorophacinone and endrin against pine voles. By this test, endrin was not as effective against meadow voles as against pine voles.

Rodenticides that can be applied as ground sprays appeal to apple growers because they already have the spray equipment and experience in using endrin. The potential of other available materials should be investigated for this use, but field trials are slow and costly. We have been studying the toxicity of endrin to pine voles and the efficacy of endrin ground sprays in controlling these rodents in the field and in the laboratory; some of our studies have suggested that endrin may act more as a tracking poison than as a toxicant ingested in the food supply. Evidence supporting this conclusion is unpublished but consists of observing that in the laboratory when endrin is mixed in the food supply at toxic levels it serves as a repellent to pine voles. Even when the voles have no other source of food, as the endrin content of the laboratory food approaches the LC50 as determined in these tests, the voles reduce the consumption of the endrin-containing food. When other food is available the effect of endrin is reduced. Yet endrin ground spray is effective against pine voles in the field.

With the thought that endrin might act as a tracking poison, Barbara C. Hackman* was able to develop a dosage-response curve by mixing endrin with soil to a known concentration, holding the animals' food uncontaminated. We have used the same methods, but with the toxicants sprayed on the soil surface. We do not know exactly how the animal acquires the toxicant; a reasonable guess seems to be that it could ingest contaminated soil particles during its vigorous grooming activities, which are so characteristic of this form, or possibly by absorption through the skin of the feet.

METHODS: The tests reported here were carried out in two sets, those of the pine vole in a laboratory on the Raleigh campus of North Carolina State University, and those of the meadow vole at the Vole Laboratory at the Mountain Horticultural Crops Research Station at Fletcher, North Carolina. The same general methods were used with some differences in details.

All animals used in the tests were adult voles, captured wild in Henderson county. With the Raleigh tests of pine voles, only adult males were used, housed three in a cage. In the Fletcher tests with meadow voles, two animals were used per cage, one male and one female. Animals were held in captivity for a minimum of 7 days before any trials.

*Then with North Carolina State University.

Table 1. Mortality of adult male pine voles exposed to three toxicants applied as ground sprays on earth substrate; 10-day exposure.

Toxicant	Exposure lbs. active per acre	Mortality
Brodifacoum	0.024	0/3
	0.049	0/3
	0.098	1/6
	0.147	0/3
	0.196	4/6
Chlorophacinone	0.18	1/3
	0.35	0/6
	0.52	2/3
	0.71	6/6
Endrin	0.94	0/3
	1.88	1/6
	2.82	1/3
	3.76	4/6
Control	0.0	0/11

All tests of ground sprays were carried out on a soil substrate in a metal box measuring 23.5 x 15 inches, 6 inches deep. Each box was fitted with a top of $\frac{1}{2}$ -inch hardware cloth; each box had a layer of about 25-30 pounds of sandy loam soil containing a moderate amount of organic matter. This soil had been sifted through a $\frac{1}{2}$ -inch hardware cloth screen to remove stones, clumps of soil and pieces of plant material. Small amounts of water were added to the soil periodically to maintain soil moisture. For food, apples were suspended above the soil surface to prevent direct contamination. A water bottle was provided. No nesting or other cover was provided to the animals, which lived on the soil substrate. Control animals were held on untreated soil in the Raleigh tests but not in those at Fletcher. Animals were observed every 24 hours or more often and body weights were measured every other day. Tests at Raleigh were terminated at the end of 10 days and at Fletcher at the end of 21 days.

All toxicants were applied mixed in water to a uniform volume of about 125 ml. This spray application was made as uniform as possible and allowed to dry overnight in the Raleigh tests, and for four hours in the Fletcher tests, before test animals were introduced. The dilute spray applications were equivalent to approximately 560 gallons per sprayed acre. In the Raleigh tests, solutions were made up from commercially available formulations of endrin and chlorophacinone, and from technical brodifacoum provided by the ICI Americas Corporation. In the Fletcher tests, the concentrations of endrin were prepared from technical

material provided by Velsicol Corporation; to allow dispersion in water the endrin was dissolved in a small quantity of xylene with 3.5 percent emulsifier.

Table 2. Mortality of adult meadow voles exposed to endrin applied as ground spray on earth substrate; 21-day exposure.

Exposure lbs. active per acre	Mortality
7.0	9/22
14.0	10/20
21.0	11/16
24.5	9/12
28.0	10/10
35.0	8/8
42.0	8/8
56.0	6/6

In fitting a dosage response curve to the mortality data from the laboratory tests, probit analysis was carried out, using the SAS system (Barr et al. 1979) with log-transformed values of the variable application rates.

RESULTS: Tables 1 and 2 show the mortality of voles exposed to ground sprays with the application rate expressed as pounds per acre of active ingredient. Table 1 shows the results for pine voles with the three rodenticides: brodifacoum, chlorophacinone, and endrin. Table 2 shows the results for meadow voles exposed to endrin. In Table 3, results are combined and restated as median lethal application rates and, where possible, compared with the rates of application on the registered labels for chlorophacinone and endrin.

DISCUSSION: For both endrin and chlorophacinone, the median lethal application rate for pine voles, as determined in the laboratory tests, was of the same order as that directed by instructions on the label, making allowance for stating both rates as per sprayed acre. We do not understand why the results from the bioassay approximate so closely the label recommendation as to application rate. If the bioassay test and the field application were exactly comparable, and we would not expect that they would be, then we would expect the median lethal dose to be something less than the effective field application rate. We need more experience in comparing the two methods of exposure before we can understand this question, but one explanation might be that in the field, some rodenticide is obtained through the food supply, a route we attempted to eliminate in our laboratory tests. Or, perhaps the spray-covered vegetation provided more surface from which the rodenticide might be picked up on the fur and ingested in grooming.

Table 3. Median lethal application rate of rodenticide ground spray in laboratory tests as compared with label rates.

Rodenticide	Species	n	Application rates as lb/acre active	
			LC50 in lab tests (95% confidence limits)	Rate specified on label
Brodifacoum	Pine vole	21	0.18 (0.12-*)	-
Chlorophacinone	Pine vole	18	0.42 (*)	0.30 ^{1/}
Endrin	Pine vole	18	3.13 (1.88-*)	3.0 ^{1/}
Endrin	Meadow vole	102	10.63 (7.05-13.64)	3.0 ^{1/}

* Unable to set limits because of heterogeneity of data.

^{1/} Label rates interpreted as pounds of active ingredient per sprayed acre. Based on area sprayed within tree drip lines, sprayed area is assumed to be 2/3 of orchard area.

Comparative results with the two species of voles suggest that endrin may not be as effective against the meadow vole as it is against the pine vole. These results are supported by observations in one orchard where endrin was reported to have been used without success, and where we found surviving animals to be meadow voles. For these tests, we obtained the test animals from other orchards. At present we do not have any stronger confirming information that endrin is not effective as a ground spray for eliminating meadow voles.

In developing this laboratory bioassay method for testing ground sprays we hope, of course, that it could serve as a less expensive screening method for investigating candidate ground-spray materials to replace endrin. Clearly, this method is no substitute for good field trials, but it has the advantage that tests can be made under controlled conditions and without special permits or clearance if nonregistered materials are used. Although these laboratory tests cost less than well-conducted field trials, they cost more, and are slower than the ordinary toxicity trials carried out in small laboratory cages.

LITERATURE CITED

Barr, A. J., J. H. Goodnight, J. P. Stall, W. H. Blair, and D. M. Chilko. 1979. SAS user's guide 1979 edition. Raleigh, NC, The SAS Institute, pp 357-360.