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2-23-1978

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Palmateer, Steve D., "Progress in Developing a Microtus Efficacy Test Method for Registration Purposes" (1978). *Eastern Pine and Meadow Vole Symposia.* Paper 99.
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PROGRESS IN DEVELOPING A MICROTUS EFFICACY TEST METHOD FOR REGISTRATION PURPOSES

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The purpose of this presentation is to document the progress that is being made in the development of a *Microtus* rodenticide efficacy test method. When finished the method will satisfy EPA requirements for a laboratory alternate diet bioassay. We have been working on the method for 2 to 3 years.

One of the biggest problems has been in attempting to combine both meadow and pine voles in a single method. Also the method should be applicable for *Microtus* toxicants designed to be applied at many use sites in the entire 50 states.

In California and Western States *Microtus* depradation can be severe in alfalfa, barley and other grain fields, truck crops such as brussel sprouts and potatoes. On the east coast vole damage is severe but not limited to orchards. The EPA apple orchards in Beltsville are heavily girdled by voles and several trees decline and die each year. To make matters worse *Microtus* are carriers of many diseases, the most important being tularemia.

Therefore designing a laboratory test method that will reveal the efficacy of a vole toxicant in all these use sites has been very difficult. I would like to briefly describe the method as it is at this time. In short the state of the art.

The apparatus used to test the voles are screened bottom all-metal cages. It has been our experience that many vole species cannot be grouped together and therefore all of them are individually caged. In one group tank test with 20 meadow voles (*Microtus pennsylvanicus*) with no toxicant present and commercial rodent feed offered *ad lib*. 7 individuals died. We attribute the deaths to fighting and cannibalism.

Laboratory temperature should be about 20 to 25° C and there should be 12 hours of artificial light per day. Water is available to the voles at all times.

The rodenticide-treated bait and the standard field rodent diet are offered to test voles in separate containers on opposite sides of the cage. There is more than enough food in each cup to supply the daily food requirements.

The standard field rodent challenge diet is composed of 50 percent (by weight) rolled oat groats, and 50 percent commercial rodent laboratory chow. The commercial rodent food was not palatable enough by its self to realistically create a challenge with the poison bait. The field rodent diet is not as palatable as the diet used to challenge

commensal rodents.

An untreated control (check) group of 20 animals is required and is offered only the field rodent diet. If more than 10% of control voles die the entire test is voided. Food consumption is not recorded for the control animals.

The test vole consumption of both poison bait and challenge diet is determined daily and returned to starting weight by addition of the given food. Every day the quantity of food consumed by each vole is recorded. Recordings should be made at the same time each day. Weighing accuracy shall be to at least the nearest half-gram. The Animal Biology Laboratory weighs all rodenticide products to the nearest tenth of a gram. Spilled rodenticide and challenge diet are recovered and weighed to establish exact food consumption data. When the food spillage has gotten wet it must be dried to original moisture content before weighing.

The position of the vole toxicant and the standard field rodent diet containers must be reversed every day to reduce any feeding position bias of the animals. There must be a free choice between the rodenticide and the challenge diet. The voles must not be stressed unduly from noise or human disturbance. The Animal Biology Laboratory maintains all test rodents in a room separate from its main laboratory to reduce stress to the test subjects.

The length of the test period is 3 days for acute (single-dose) rodenticides and 15 days for anticoagulants. Dead voles are removed daily. All rodenticides are removed at the end of the test period leaving only the standard field rodent bait. No further weighing of food consumption is required.

Observation is maintained on surviving voles for 5 days following the test period. Any deaths encountered during this time period is attributed to the rodenticide. Some toxicants may require up to a 10 day post observation period. Sound rational for this extended post-test observation period would have to be presented. It is possible some of the potent anticoagulants tested for 3 days could justify a 10 day post-test period.

A vole toxicant (either single or multidose) would be considered satisfactory if a minimum of 90 percent mortality of test animals is obtained. Vole baits with exceptional safety characteristics to humans and other nontarget animals with a high degree of usefulness in special control situations may have a parameter of efficacy reduced to 80 percent.

Some of the problems encountered by the Animal Biology Laboratory in using this method are: 1. Mortality in control animals, 2. lack of a suitable nest container in each cage, 3. food dish may inhibit easy access to rodenticide and challenge diet, and 4. lack of a large uninterrupted supply of voles.

The Animal Biology Laboratory has or will in the near future test the following listed active ingredients in various concentrations on pine and meadow voles:

1. Strychnine Alkaloid
2. Warfarin
3. Sodium Fluoroacetate
4. brodifacoum
5. Zinc Phosphide
6. diphacinone
7. chlorophacinone
8. DLP-787

The use of generic or brand names does not imply endorsement by the Federal government. Finally, I would like to acknowledge the cooperation of Dr. Ross Byers in the development of this test method.