

STANDARD HOUSE MOUSE ANTICOAGULANT TRACKING POWDER EFFICACY

Revision No. 9

LABORATORY TEST METHOD

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OPP Designation: 1.212 (1-1-75)

1. Scope

1.1 This method is designed to determine effectiveness of anticoagulant tracking powder rodenticide products used for control of house mice. This method is applicable in connection with registration and enforcement procedures under the Federal Insecticide, Fungicide, and Rodenticide Act, as amended. The conduct of, reporting of, and recordkeeping for studies conducted according to this method must conform with the U.S. Environmental Protection Agency's "Good Laboratory Practice Standards" (40 CFR, Part 160).

2. Test Animals

2.1 All mice used in this test shall be house mice (Mus musculus), wild-type (wild-caught or from a wild mouse colony) or albinos (Swiss Webster strain preferred). They shall be healthy, active, sexually mature, and fall within the following weight classes in grams within seven days prior to start of the test:

	<u>Minimum</u>	<u>Maximum</u>	<u>Maximum acceptable differences in average weights between sexes</u>
Laboratory mice	15	35	5
House mice	10	25	3

2.2 Ectoparasite control with registered insecticide (or acaricide) products labeled for use on laboratory rodents is permissible if applied externally to both test and control animals not less than seven days prior to start of test, if applied at rates not exceeding those permitted by the registered label, and if the pesticide used is not known or believed to potentiate the effects of the rodenticide in the product being tested.

3. Apparatus

3.1 Test apparatus consists of two screened-bottomed cages measuring at least 38.1 cm by 45.7 cm by 22.9 cm (15 by 18 by 9 in.) high and connected with one another by two square hardware cloth tunnels, 91.4 cm (36 in.) long with sides, top, and bottom all measuring 11.5 cm (4 1/2 in.) The tunnels are parallel and spaced 10.2 cm (4 in.) apart. A galvanized or stainless steel removable pan, 30.5 cm (12 in.) long by 11.4 cm (4 1/2 in.) wide with a 6 mm (1/4 in.) lip on both sides is centered in each tunnel. Each cage should contain shelters. Empty soup or beverage cans, with one end removed, slightly flattened to prevent rolling, have been found satisfactory for this purpose. Use at least two cans per five mice.

3.2 Both of the interconnected cages must contain one food container for every five mice in the subgroup. Metal or ceramic feeders, designed so that test mice may not nestle or wallow in diet, should be used.

3.3 Each of the interconnected cages must contain one no-drip waterer fitted with ball-type watering tubes. Provide one waterer for every 5 mice in the subgroup.

4. Pretest Holding Conditions

4.1 All mice used in this test method must be held, group-caged with sexes separate, for observation in the laboratory for a period of at least one and not more than four weeks prior to testing. During the last seven days of this period, mice shall be held under laboratory conditions (i.e., temperature, humidity, lighting, etc.) comparable to those of the animal testing room if not actually in the testing room. The test animals must not be fasted prior to testing. Water and a commercial mouse diet must be available to them at all times.

5. Holding and Test Conditions

5.1 Temperature	20 to 25° C. Strong air currents from heaters or air conditioners shall not blow directly onto test animals.
Relative humidity	50 to 55%.
Light	12 h artificial light per day, not to exceed 2153 lx (200 ft candles) at cage location. Total reversing of the natural photoperiods of the test animals by timed lighting is not recommended.

6. Procedure

6.1 A test group consists of a minimum of 20 mice (10 males, 10 females) group-caged in single-sex subgroups of 5 or 10 animals. For each test or series of tests conducted at the same time on the same strain, include one untreated control test group of 20 mice (10 males, 10 females), caged in the same manner as the group(s) to be exposed to toxic tracking powder. Acclimate all animals to test conditions for three days prior to exposure to toxicant, immediately following pretest holding period (4.1).

6.2 Fill food containers (3.2) in each of the interconnected cages daily with a commercially available laboratory mouse diet. Provide at least 15 grams of feed per animal per day in each of the interconnected cages.

6.3 Fill waterers (3.3) daily to provide at least 200 ml of tap water for every five subjects in the subgroup.

6.4 Place test subgroups of five or ten animals of the same sex into separate apparatuses. Each study group is to be divided into either a subgroup of 10 males and one of 10 females or into two subgroups of five males each and two subgroups of 10 females each.

6.5 Commencing five days after the introduction of mice in the testing apparatus, dust one of the removable steel pans with the tracking powder (the same tunnel throughout the test) each day according to instructions given on the label. As most product label direct users to to dust lightly, 9 grams of the dusting powder should be used daily.

6.6 Maintain a daily record of activity across the powder on the treated plate. Cover all previous signs of activity with fresh powder after the daily reading has been taken.

6.7 Low-current electric sensing devices and activity counters may be installed in each tunnel to record mouse movement through both the treated and untreated tunnels.

6.8 Animals on test should not be subjected to undue or unnecessary stress from noise or human activities (i.e., movement). Human activity within the animal test room shall be minimal.

7. Test Period

7.1 Maintain test period for 15 days, counted from the day when tracking powder was introduced.

7.2 Remove dead mice daily, or more frequently as observed.

7.3 Remove, thoroughly clean, and replace the tracking powder removable pan in the tunnel after the test period.

7.4 This laboratory efficacy test should be replicated at least once.

8. Test Period Follow-Up

8.1 Maintain observation on surviving test and control group mice for a minimum of five days following test (powder exposure) period.

8.2 Continue feeding commercial rat and mouse diet as in 6.2.

8.3 Describe unusual activities of test animals in tunnels and cages in report of test and posttest periods.

9. Calculation and Evaluation of Results

9.1 Record date, weight, and sex of each mouse dying during the study and of survivors in test and control groups. Retain original laboratory test records for future reference. Report all data collected including initial and final weights of test subjects. Include copies of all "raw" data sheets and type summaries of test results

9.2 The product is considered satisfactory if mortality of at least 90% is obtained in test animals.

9.3 The test report must include a report of a chemical analysis of the test product, conducted using methods that are acceptable to the U. S. Environmental Protection Agency.