

STANDARD NORWAY RAT/ROOF RAT ANTICOAGULANT

Revised:

7-23-74

1-1-75

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8-15-80

6-19-91

TRACKING POWDER EFFICACYLABORATORY TEST METHOD

OPP Designation: 1.205 (2-25-74)

1. Scope

1.1 This method is designed to determine effectiveness of anticoagulant tracking powder rodenticide products used for control of Norway rats or roof rats. It 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 rats used in this test shall be Norway rats (Rattus norvegicus), wild-type (wild-caught or from a wild-type Norway rat colony) or albinos (Wistar strain preferred), or wild-type roof rats (R. rattus). Subjects shall be healthy, active, sexually mature, and fall within the following weight classes in grams within seven days prior to start of test:

	<u>Minimum</u>	<u>Maximum</u>	<u>Maximum acceptable differences in average weights between sexes</u>
Laboratory rats	150	300	50
Norway rats	150	400	65

2.2 Ectoparasite control with registered insecticide (or acaricide) products labeled for use on laboratory rats 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 anticoagulant rodenticides.

3. Apparatus

3.1 Test apparatus consists of two 568 liter (150 gal) stock watering tanks (with tops cut away to facilitate observation, feeding, and watering of test rats) connected by two square hardware cloth tunnels, 914 mm (36 in.) long with 114 mm (4 1/2 in.) sides, top, and bottom. The tunnels are parallel and spaced 1016 mm (40 in.) apart. A galvanized or stainless steel removable pan, 305 mm (12 in.) long by 102 mm (4 in.) wide with a 6 mm (1/4 in.) lip on both sides is centered in each tunnel. Each tank should contain an open-bottom metal nest box approximately 36 cm X 36 cm X 10 cm (14 in. X 14 in. X 4 in.) in size.

3.2 Each tank should contain metal or ceramic feeders designed so that test rats may not nestle or wallow in diet.

3.3 Each tank should contain 1000 cc chicken founts. Automatic waterers are not recommended.

#### 4. Pretest Holding Conditions

4.1 All rats used in this test method must be held, sexes separate, group caged, 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, rats 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 (3.3) and a commercial rat and 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 Fill food containers (3.2) in each tank with a commercial laboratory rat and mouse diet. Provide at least 40 grams per animal per day in each tank. Replenish food containers as needed. The containers must be identical in type and size. The food offered in each container should be equal and consistent throughout the test. If food becomes fouled by urine or feces, replace all food in each container.

6.2 Fill waterers (3.3) in each tank with tap water to provide each animal with at least 60 ml of water per day. Replenish waterers daily to capacity.

6.3 The test animals must have free and equal access to both tanks (3.1) through the tunnels.

6.4 Test groups consist of 20 rats (10 males and 20 females). All 20 animal may be tested in one two-tank enclosure. Alternatively, and preferably, put all 10 males in one two-tank enclosure and all 10 females in a second, similarly constructed two-tank enclosure. For each test or

series of tests conducted at the same time and on the same species, include one similarly housed control group of 20 rats (10 males, 10 females).

6.5 Commencing five days after the introduction of rats 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 labels specify to dust lightly, 9 grams of the dusting powder may 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 rat movement through both the treated and untreated tunnels.

6.8 Animals on test shall 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 introduction of tracking powder).

7.2 Remove dead rats daily.

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

7.4 This test should be replicated at least once.

## 8. Test Period Follow-Up

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

8.2 Continue feeding commercial rodent laboratory diet as in 6.1.

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

## 9. Calculation and Evaluation of Results

9.1 Record date, weight, and sex of each rat dying during the test 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 typed summaries of test results.

9.2 The product is considered satisfactory if a mortality of at least 90% is obtained among the test group animals during the test and posttest periods and if no more than 10% of control-group subjects die during the same period of time.

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