Revision No. 9
Revised:
1-1-75
9-1-76
2-21-78
8-15-80
6-21-91
9-27-91

STANDARD HOUSE MOUSE ACUTE LIQUID BAIT

LABORATORY TEST METHOD

OPP Designation: 1.208

1. Scope

1.1 This method is designed to determine effectiveness of acute rodenticide products used to make liquid baits for controlling 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 test:

	Minimum	Maximum	Maximum acceptable differences in average weights between sexes
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 The mice may be housed individually or in single-sex groups of 5 or 10 mice per group. Mice should be placed in solid-bottom all-metal cages designed to hold laboratory mice or in specially constructed or modified cages suitable for maintaining house mice for this type of study. If mice are housed singly, cages must have a botton surface area of at least 500 cm^2 (0.538 ft^2). If mice are group-caged, each enclosure must have a bottom surface area of at least 2,000 cm^2 (2.15 ft^2).

3.2 If subjects are group-caged, provide shelters in both the test and control cages. Empty soup or beverage cans, with one end removed, slightly flattened to prevent rolling, have been found satisfactory for this purpose. Use two cans for every five mice in the enclosure.

3.3 Metal or ceramic feeders, designed so that test mice may not nestle or wallow in diet, should be used.

3.4 Graduated 50-ml or 100-ml no-drip waterers fitted with ball-type watering tube should be used. Automatic or open-cup type waterers are not recommended.

4. Pretest Holding Conditions

4.1 All mice used in this test method must be held, sexes separate, for observation in the laboratory for a period of at least two 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. Do not use the standard EPA challenge diet for pretest feeding.

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) individually-caged or 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 bait. Acclimate all animals to test conditions for three days prior to exposure to toxicant, immediately following pretest holding period (4.1).

6.2 For individually-caged subjects, provide one or two feeders filled daily with a commercially available laboratory rat diet. For group-caged subjects, use two or more feeders per enclosure. Provide at least 15 grams of feed per animal per day.

6.3 For individually-caged subjects, provide two 50- or 100-ml graduated no-drip waterers (3.4). Fill one waterer with toxic bait solution and the other with tap water. For group-caged subjects, provide each treated test group with a minimum of four 100-ml graduated no-drip waterers (3.4), half of which are to be filled with tap water and the other half to be filled with

the toxic liquid bait formulation diluted with tap water according to the mixing directions on the product's label. Position waterer relative to food sources and other features so that the cage environment does not clearly favor any waterer position over any other. Replenish both liquids as necessary so that no waterer becomes less than approximately one-third filled. Reverse position of the waterers daily.

6.4 Provide control-group cages with waterers of the same number and type that are used for test-group cages. All waterers provided to control-group animals must be filled filled with tap water. Replenish as necessary so that no waterer becomes less than approximately one-third filled.

6.5 Each day, record the total quantity of each liquid consumed during the preceding 24 h for both the test and control groups.

6.6 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 2 days.

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

7.3 Remove toxicant waterers at the end of the 2-day test period, leaving and maintaining tap water waterers.

7.4 More than a 10% mortality in the control group negates the test, even if a 100% mortality had been achieved in the test group.

7.5 This test should be replicated at least once.

8. Test Period Follow-Up

8.1 Maintain observation on surviving mice in test and control groups for a minimum of five days following test period.

8.2 Continue commercial rodent laboratory diet as in 6.2.

8.3 Describe unusual activities of test and control mice 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 test and of survivors in both the test and control groups, and amount of toxic and nontoxic liquids consumed during the test and posttest periods. 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 as well as typed numerical summaries of test results.

9.2 The product is considered satisfactory if a minimum mortality of 90% of test animals is obtained during the bait-exposure and post-exposure observation periods and if no more than 10% of control-group subjects die during the study.

9.3 The test report must include reports of chemical analyses of the test bait solution and the tap water for the active ingredient claimed to be in the test product. These tests must be conducted using methods that are acceptable to the U. S. Environmental Protection Agency.