

1. CHEMICAL: Brodifacoum
2. FORMULATION: Formulated (0.005% A.I) Pellet bait
3. CITATION: Marsh, Rex E. and Howard, Walter E. (1978). Secondary toxicity Hazards Tests of Brodifacoum to Raptors (preliminary Report not for distribution.) Submitted by ICI Americas Inc., EPA Acc. #245704. Submitted July 22, 1981.
4. REVIEWED BY: Russel Farringer
Wildlife Biologist
EEB/HED
5. DATE REVIEWED: 10/29/81
6. TEST TYPE: Secondary Toxicity
 - A. TEST SPECIES: Golden Eagle (Aquila chryseatos), Red-tailed hawks (buteo borealis colurus), red-shouldered hawks (Buteo lineatus)
7. REPORT RESULTS:

All four eagles tested survived, although what appears to be anticoagulant-related symptoms occurred in three out of four birds. All four Red-tailed and one of two Red-shouldered hawks died during the course of the test.
8. REVIEWER'S CONCLUSIONS: ✓ While this study had some short comings the data could still be of value in a hazard assessment. Because of the nature of this study and the study design parts of the report will be exert for future reference. Since this is a preliminary report and the researcher raise some important points on its validity, we will use the information contained in it as indicative and not absolute.

Materials and Methods

Test Procedure

(Excerpted from text of Report)

In order to be able to replicate these tests in our own laboratory, and so the same could be done in other research centers, we established a set of rather definite parameters for preparing the rodenticide-treated rodents and how they were fed to the raptors. These procedures follow.

1. Use only wild rodents, as opposed to laboratory strains, in the secondary-hazard feeding tests.
2. Have the treated baits to be offered to the test rodents contain concentrations of rodenticide which are consistent to expected use practices.
3. Cage the test rodents individually and offer them the bait treated with the anticoagulant (brodifacoum) for 3 consecutive days in a no-choice situation, i.e., with no other food available.
4. Record daily the amount of bait consumed in the 3-day test and calculate the total mg of rodenticide consumed by each rodent (multiply the amount of bait consumed by the concentration of the toxicant in the bait).
5. Following this 3-day feeding period, put the rodents on normal laboratory diet and observe them daily (for 7 days with rats and 9 days with mice) and record day of death.
6. Only those rats which succum (to the rodenticide) during days 4 through 10 (days 4 through 12 for house mice) following the first day of the 3-day feeding are to be used in the secondary hazards study. The reason for only using test rodents that died during these prescribed number of days is to facilitate determining the amount of brodifacoum they have taken in, and those living longer before dying may lose too much of the rodenticide before dying.
7. All rodents which are atypical in the amount of bait consumed or die outside the established time parameters are to be discarded.
8. Observe all test rodents several times daily; when death occurs place the animals and a record of all pertinent data concerning that animal in a plastic bag which is sealed and then frozen at 24°C (-14°F) until needed for the secondary hazards study.
9. About 16 hours before the rodenticide-laden rodents are to be fed to the raptors, remove them from the freezer and place them on trays to thaw at room temperature, generally overnight, so that partly frozen carcasses, which would influence "normal" feeding behavior, are never offered to the raptors.

10. For pre- and posttreatment feeding of raptors, wild rodents of the same species are euthanized with CO₂ and stored frozen until needed.

11. A detailed record of all data is prepared about each rodent and a copy is retained with each carcass.

Results & Discussion

- (A) "It is recognized that the rodents described in the above no-choice feeding test procedures have probably 1) ingested what might be considered the maximum amount of rodenticide possible over a 3-day period; (2) that this amount is many times the quantity of brodifacoum needed to be lethal to the rodents; and 3) it is many times in excess of what might be consumed under natural field baiting operations." pg 3 of Report.

Report no: CTL/P7462 Brodifacoum: Absorption, Excretion and Tissue Retention in the Rat by H. Bratt, & Pamela Audson (1979) ICI. Lab.

'Their results showed that absorption may be a saturable process with fecal excretion increasing rapidly after saturation.'

In relationship to the above enumerated statements, we can agree with one and two. Statement three is unsubstantiated. If the bait was the easiest and most readily available to the rodent then one would assume that the rodent could consume large amounts of the bait.

- (B) "Prior to going on to the test, each bird was maintained on fresh-frozen and then thawed carcasses of day-old chicks obtained from a hatchery by the Raptor Center." (pg 3)

"Prior to testing, the raptors were being fed as many day-old baby chicks as they would consume. How much vitamin K this provided the birds is of particular concern to this study." (pg 5)

"At [another] raptor center, it was concluded that the birds were deficient in vitamin K; and upon supplementing their diet with vitamin K, much greater resistance to anticoagulents resulted in subsequent testing." (pg 5)

Since the Raptor were fed entire baby chicks they could have received large doses of vitamin K than they would normally receive in the wild. No testing was done that would have determined if this biased the results of this test.

The attached tables give mortality data and dose level.

Table 1. Days to death, and amount of .005% brodifacoum (r.p.581) treated EPA rodent diet consumed by the wild Norway rats (*Rattus norvegicus*) fed to the golden eagles (*Agulta chrysaetos*) in Table 5.

| Bird No. | Rodent No. | Rodent wt. (kg)* | Toxic bait consumed (g) | | | Amount consumed | | Time to death (days) |
|----------|------------|------------------|-------------------------|-------|-------|-----------------|------------|----------------------|
| | | | Day 1 | Day 2 | Day 3 | Bait (g) | PP581 (mg) | |
| GE-4 | D-18 | .335 | 30.1 | 27.1 | 15.2 | 72.4 | 3.62 | 6 |
| | D-1 | .353 | 20.4 | 22.7 | 27.6 | 70.7 | 3.54 | 7 |
| | D-10 | .265 | 1.4 | 30.7 | 35.1 | 67.2 | 3.36 | 8 |
| | F-1 | .428 | 38.5 | 36.7 | 33.1 | 108.3 | 5.42 | 7 |
| GE-5 | D-9 | .250 | 23.3 | 20.6 | 30.9 | 74.8 | 3.74 | 8 |
| | D-14 | .256 | 23.1 | 21.9 | 26.1 | 71.1 | 3.56 | 6 |
| | D-7 | .306 | 12.9 | 24.9 | 30.0 | 67.8 | 3.39 | 7 |
| | D-5 | .474 | 23.7 | 26.0 | 33.2 | 82.9 | 4.15 | 7 |
| GE-7 | D-8 | .271 | 20.0 | 22.3 | 32.1 | 74.1 | 3.71 | 7 |
| | D-19 | .371 | 3.7 | 32.7 | 33.8 | 70.2 | 3.51 | 7 |
| | D-6 | .297 | 17.3 | 23.6 | 25.9 | 66.8 | 3.34 | 6 |
| | D-17 | .366 | 36.8 | 31.7 | 39.3 | 107.8 | 5.39 | 8 |
| GE-8 | F-5 | .269 | 30.1 | 22.0 | 24.7 | 76.8 | 3.84 | 8 |
| | D-2 | .353 | 17.6 | 22.4 | 31.7 | 71.7 | 3.59 | 6 |
| | F-8 | .253 | 22.2 | 21.8 | 24.8 | 68.8 | 3.44 | 9 |
| | F-7 | .386 | 24.0 | 26.4 | 26.5 | 76.9 | 3.85 | 8 |

*Rodents are listed in the order they were fed to the golden eagles.

Table 2. Days to death, and amount of .005% brodifacoum (PP581) treated EPA rodent diet consumed by the wild Norway rats (Rattus norvegicus) fed to the western red-tailed hawks (Buteo borealis calurus) in Table 6.

| Bird No. | Rodent No. | Rodent wt. (kg)* | Toxic bait consumed (g) | | | Amount consumed | | Time to death (days) |
|----------|------------|------------------|-------------------------|-------|-------|-----------------|------------|----------------------|
| | | | Day 1 | Day 2 | Day 3 | Bait (g) | PP581 (mg) | |
| RT 58 | D-11 | .228 | 15.9 | 18.9 | 14.7 | 60.1 | 3.01 | 7 |
| | D-12 | .237 | 9.3 | 17.5 | 26.8 | 53.6 | 2.68 | 9 |
| | D-15 | .315 | 0.3 | 17.7 | 32.3 | 50.3 | 2.52 | 8 |
| | F-13 | .300 | 22.6 | 22.7 | 20.5 | 41.4 | 2.07 | 10 |
| RT 70 | D-13 | .265 | 11.7 | 19.5 | 28.9 | 60.1 | 3.01 | 8 |
| | D-20 | .204 | 16.0 | 17.0 | 21.4 | 54.4 | 2.72 | 8 |
| | F-12 | .170 | 17.4 | 16.8 | 15.6 | 49.8 | 2.49 | 8 |
| | F-4 | .157 | 5.5 | 17.6 | 17.7 | 40.8 | 2.04 | 7 |
| RT 107 | D-3 | .284 | 15.8 | 20.9 | 24.0 | 60.7 | 3.04 | 8 |
| | D-16 | .290 | 4.9 | 21.8 | 32.9 | 59.6 | 2.98 | 7 |
| | F-3 | .198 | 18.8 | 14.1 | 11.9 | 44.8 | 2.24 | 9 |
| | F-11 | .180 | 7.7 | 16.0 | 16.0 | 39.7 | 1.99 | 10 |
| RT 116 | F-9 | .184 | 22.5 | 21.0 | 18.6 | 62.1 | 3.11 | 6 |
| | F-6 | .161 | 17.8 | 18.5 | 19.2 | 55.5 | 2.78 | 6 |
| | F-15 | .156 | 19.7 | 17.0 | 14.6 | 51.3 | 2.57 | 6 |
| | F-14 | .150 | 15.3 | 11.2 | 7.6 | 34.1 | 1.71 | 7 |

*Rodents are listed in the order they were fed to the western red-tailed hawks.

Table 3. Days to death, and amount of .005% brodifacoum (PP581) treated EPA rodent diet consumed by the wild house mice (Mus musculus) fed to the red-shouldered hawks (Buteo lineatus) in Table 7.

| Bird No. | Rodent No. | Rodent wt. (kg)* | Toxic bait consumed (g) | | | Amount consumed | | Time to death (days) |
|----------|------------|------------------|-------------------------|-------|-------|-----------------|------------|----------------------|
| | | | Day 1 | Day 2 | Day 3 | Bait (g) | PP581 (mg) | |
| RS 6 | H-6 | .0179 | 3.5 | 3.6 | 4.1 | 11.2 | .56 | 10 |
| | H-11 | .026 | 3.3 | 3.8 | 4.3 | 11.4 | .57 | 12 |
| | H-13 | .0222 | 3.2 | 4.2 | 3.7 | 11.1 | .56 | 10 |
| | H-14 | .0237 | 1.8 | 3.4 | 4.6 | 9.8 | .49 | 5 |
| | H-19 | .0188 | 2.2 | 2.9 | 4.2 | 9.3 | .47 | 10 |
| | H-33 | .0156 | 3.4 | 3.7 | 3.5 | 10.6 | .53 | 11 |
| | H-12 | .0146 | 3.6 | 3.3 | 2.5 | 9.4 | .47 | 6 |
| | H-21 | .0168 | 1.5 | 2.3 | 3.7 | 7.5 | .37 | 5 |
| | H-22 | .0158 | 1.4 | 2.7 | 3.3 | 7.4 | .37 | 9 |
| | H-4 | .0206 | 4.8 | 4.3 | 4.1 | 13.2 | .66 | 7 |
| | H-26 | .0151 | 1.4 | 2.1 | 3.0 | 6.5 | .33 | 7 |
| | H-29 | .0169 | 1.7 | 2.2 | 3.0 | 6.9 | .35 | 9 |
| RS 10 | H-31 | .0158 | 3.7 | 4.0 | 3.4 | 11.1 | .56 | 12 |
| | H-35 | .0154 | 3.8 | 4.2 | 3.5 | 11.5 | .58 | 10 |
| | H-38 | .0184 | 4.0 | 3.6 | 3.5 | 11.1 | .56 | 7 |
| | H-18 | .0237 | 3.1 | 2.9 | 3.8 | 9.8 | .49 | 6 |
| | H-32 | .0199 | 3.5 | 3.8 | 3.2 | 10.5 | .53 | 10 |
| | H-39 | .0149 | 2.9 | 3.5 | 3.0 | 9.4 | .47 | 9 |
| | H-16 | .0163 | 2.0 | 3.6 | 3.6 | 9.2 | .46 | 9 |
| | H-24 | .0154 | 1.7 | 2.5 | 3.1 | 7.3 | .37 | 7 |
| | H-25 | .0145 | 1.9 | 2.7 | 3.1 | 7.7 | .39 | 8 |
| | H-7 | .0192 | 4.6 | 5.0 | 3.9 | 13.5 | .68 | 9 |
| | H-23 | .0153 | 0.5 | 2.6 | 3.4 | 6.5 | .33 | 9 |
| | H-30 | .0152 | 1.8 | 2.5 | 2.5 | 6.8 | .34 | 12 |

*Rodents are listed in the order they were fed to the red-shouldered hawks.

Table 5. A secondary-feeding test with golden eagle (*Haliaeetus auratus*), including pre- and postfeeding periods and the number of additional days the birds were observed for symptoms, which were fed wild honey bees (*Hyalophora*) that had died from eating red-tail bait containing .0025 brodifacoum (77261). See Table 1 for details about these tests. The actual amount of 77261 consumed by the eagles is unknown because of detoxification and other factors but the maximum potential amount that could be in the rats eaten by the eagles is shown.

| Bird No. | Bird wt. (kg) | 2-day pretest feeding period Total red-tail consumed (g) | 4-day pretest feeding period Total red-tail consumed (g) | 4-day pretest feeding period Total no. of rats consumed (g) | Maximum observed consumed mg/kg | Maximum observed consumed mg/kg | 2-day pretest feeding period Total red-tail consumed (g) | Additional observation period no. of days | Test symptoms observed yes/no | Normalizing period |
|----------|---------------|--|--|---|------------------------------------|------------------------------------|--|---|----------------------------------|-----------------------|
| GE-4 | 3.459 | 458 | 1 | 4 | 946 | 15.93 | 4.01 | 27 | no | no |
| GE-5 | 3.147 | 516 | 1 | 4 | 946 | 14.83 | 4.71 | 27 | yes | 12 (external blood) |
| GE-6 | 4.481 | 547 | 1 | 4 | 1093 | 13.59 | 3.45 | 27 | yes | 9 (salivary) |
| GE-7 | 3.457 | 639 | 1 | 4 | 1149 | 14.71 | 4.08 | 27 | yes | 11 (external blood) |

Table 6. A secondary-ventilation test with western red-tailed hawks (*Buteo borealis calurus*) including pre- and postfeeding periods and the number of additional days the birds were observed for symptoms, which were fed with honey rats (*Reithrodontomys*) that had died from eating rodent bait containing .005 levamisole (F7501). See Table 8 for details about these rats. The actual amount of F7501 consumed by the birds is unknown because of denatification and other factors but the minimum potential amount that could be in rats eaten by the birds is shown.

| Bird No. | Bird wt. (kg) | 2-day pretest feeding period | | 4-day pretest feeding period | | Residue chemical mg/kg | 2-day pretest feeding period | | Additional observation no. of days | Style EXAMINED yes/no | Style EXAMINED yes/no | Style EXAMINED yes/no |
|----------|------------------|------------------------------|-----------------------|---|---|---------------------------|------------------------------|-----------------------|---------------------------------------|--------------------------|--------------------------|--------------------------|
| | | Total rodents consumed | Total consumed (g) | Meaning of Total no. of rodents fed daily consumed | Total no. of rodents consumed (g) | | Total rodents consumed | Total consumed (g) | | | | |
| 01-16 | 1.516 | 3 | 322 | 3 | 304 | 10.37 | 3 | 310 | 11 | yes | 6 (external blood) | yes |
| 01-19 | 1.347 | 3 | 371 | 1 | 670 | 10.36 | 3 | 342 | 10 | yes | 8 (external blood) | yes |
| 01-109 | 1.307 | 3 | 303 | 1 | 672 | 10.34 | 3 | 323 | 3 | yes | 7 (external blood) | yes |
| 01-116 | 1.007 | 3 | 319 | 1 | 390 | 10.15 | 3 | 376 | 3 | yes | 7 (external blood) | yes |

Table 7. A secondary-territory test with red-shouldered hawks (*Buteo lineatus*) including pre- and postfeeding periods and number of additional days the birds were observed for symptoms, which were fed wild house mice (*Reithrodontomys*) that had died from eating rodent bait containing 0.01% brodifacoum (P7501). See Table 2 for details about table mice. The actual amount of P7501 consumed by the birds is unknown because of detoxification and other factors but the maximum that could be in the mice eaten by the birds is shown.

| Bird no. | Bird wt. (kg) | 3-day pretest feeding period | | 4-day pretest feeding period | | Maximum chemical | | 3-day pretest feeding period | | Additional observation | | Table symptoms observed | |
|----------|---------------|------------------------------|--|------------------------------|--|------------------|-------|------------------------------|--|------------------------|--------|-------------------------|-------------------------|
| | | Actual amount consumed (g) | Percent of total no. wt. of rodents fed daily consumed (g) | Actual amount consumed (g) | Percent of total no. wt. of rodents fed daily consumed (g) | mg | mg/kg | Actual amount consumed (g) | Percent of total no. wt. of rodents fed daily consumed (g) | Period | Period | Period | Period |
| W-6 | 570 | 9 | 118 | 2 | 12 | 164 | 5.72 | 10.40 | 9 | 119 | 27 | yes | 0 (blood in droppings?) |
| W-10 | 400 | 9 | 134 | 2 | 12 | 168 | 5.72 | 0.41 | 9 | 136 | 11 | yes | 10 (external blood) |