GUIDELINE NUMBER: 72-1 (d)


REASON FOR SUBMISSION:
FIFRA '88 Reregistration.

RESULTS-
Valid _______  Invalid _______  Supplemental  X

GUIDELINE-
Satisfied ______  Partially Satisfied  Not Satisfied  X

DISCUSSION:
The LC_{50} was calculated with two sets of measured concentrations that were done in two series. Since Brodifacoum degrades, the calculation based upon the nominal concentrations is not acceptable.

There were "On" and "Off" measurements of the concentration:

'ON' measured immediately after test concentration is prepared.
'OFF' measured 24 hours after preparation.

Because of the phrasing of these definitions, EEB infers that the chemical analysis was not done on aquarium water, but was done on the stock solutions instead. This is inappropriate because of the changes in the concentrations that may be brought about by the fishes' metabolism.

ICI never calculates the LC_{50} based upon measured concentrations, but it does offer a table that would allow the calculation to be made using the "OFF" data. The "OFF" data is more appropriate than the "ON" data, but, if variation is small, means could be used.

Little explanation is given for the two "series" of groups. "Series II" appears to have been designed to "Fill-in" gaps in the range of concentrations used. The dates of the test (March 6 to 19) lead EEB to infer that the test were conducted separately then their results were combined in order to calculate the LC_{50}.

EEB has made it's calculations by using the "OFF" data and combining the two series. The study could be upgraded to "Core" by explaining the points discussed here and/or recalculating.
the data. Since $LC_{50}$s should be reported as "percent active ingredient," EEB has converted the ICI's figures to %ai.

CONCLUSIONS: Change the classification to- "Supplemental" $LC_{50} = 0.02$ mg/kg (0.01 - 0.03), NOEL < 0.01 mg/kg.

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Date: Jan 8, 1991

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Signature: [Signature]
Date: 1-7-91
| FORMULATION: | JFU 5074 |
| % a.i. | 0.25% |
| SC # | brodifacoum |
| CHEMICAL NAME | |
| VALIDATOR: | Larry Turner |
| DATE: | April 16, 1979 |
| TEST TYPE: | Fish acute 96 hour LC₅₀ Rainbow Trout |
| TEST ID.# | ES-J1 |


**RESULTS:** Rainbow trout 96 hour LC₅₀ - 17.36 mg/l (95% c.i. 15.86-19.01 mg/l). No mortality occurred at the 3 lowest levels up to 10.0 mg/l; 100% mortality occurred at the 5 highest concentrations of 24.0 mg/l and up. Toxic symptoms included swimming at the surface, weakness, and darkened coloration. There was no observed effect on controls or fish treated at 4.2 and 5.6 mg/l. (See note at end for values based on measured concentrations.)

**VALIDATION CATEGORY:** Supplemental (core for formulated Product)

**CATEGORY RATIONALE:** Although two series of dose levels were run at separate times, there appeared to be no intent to hide this fact. Statistical analyses showed that the LC₅₀ was approximately the same for either series or for the combination. The test appeared to meet or exceed all other standards and protocol recommendations except that the formulated product was tested.

**CATEGORY REPAIRABILITY:** No repair is possible, although this test can be considered core if a test on the formulated product is required.
ABSTRACT: Rainbow trout were exposed for 96 hours to formulation JFU 5074 of brodifacoum in two series of concentrations, as given below.

<table>
<thead>
<tr>
<th>Series</th>
<th>Date Started</th>
<th>Concentration</th>
<th>96-Hour Mortality</th>
</tr>
</thead>
<tbody>
<tr>
<td>I</td>
<td>3/6/78</td>
<td>75</td>
<td>10/10</td>
</tr>
<tr>
<td></td>
<td></td>
<td>56</td>
<td>10/10</td>
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<td></td>
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<td>32</td>
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<td>18</td>
<td>6/10</td>
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<td></td>
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<td>10</td>
<td>0/10</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Control</td>
<td>0/10</td>
</tr>
<tr>
<td>II</td>
<td>3/10/78</td>
<td>42</td>
<td>10/10</td>
</tr>
<tr>
<td></td>
<td></td>
<td>24</td>
<td>10/10</td>
</tr>
<tr>
<td></td>
<td></td>
<td>15.5</td>
<td>2/10</td>
</tr>
<tr>
<td></td>
<td></td>
<td>5.6</td>
<td>0/10</td>
</tr>
<tr>
<td></td>
<td></td>
<td>4.2</td>
<td>0/10</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Control</td>
<td>0/10</td>
</tr>
</tbody>
</table>

Because of degradation and/or precipitation of the test material, concentrations were renewed each day. Fresh concentrations were measured at 67-120% of nominal, while just prior to renewal concentrations were measured at 42-82% of nominal.

Procedures very closely followed Stephan (USEPA, 1975), except that the formulated product was tested, water temperature was 13±1°C, and two different dose series were tested at different times. Statistical analysis was conducted according to the Finney probit method, combining both of the dose series. When checked on the EEB calculator, the data would not run with all 0% and 100% mortality levels included. When the lowest 0% and the three highest 100% mortality levels were excluded, an LC$_{50}$ value of 17.31 mg/l was obtained, with an acceptable chi square value. Each of two separate series was analyzed according to the Spearman-Karber method because the individual series had only one partial mortality level each. Comparable LC$_{50}$ values of 16.75 mg/l and 17.75 mg/l were obtained for Series I and II, respectively.

NOTE: In accession 237703, additional information on this test was supplied. The LC$_{50}$ values for 24, 48, 72, and 96 hours were calculated, apparently by probit analysis, based on (a) measured fresh concentrations, (b) measured concentrations just prior to renewal, and (c) mean values of the above two. These 96-hour values are given below.

Rainbow 96-hour LC$_{50}$ (.25%) = (2) 12.3 ppm, (b) 8.7 ppm, (c) 10.6 ppm
<table>
<thead>
<tr>
<th>5.17/10</th>
<th>4.47/10</th>
<th>3.97/10</th>
<th>2.87/10</th>
<th>2.17/10</th>
</tr>
</thead>
<tbody>
<tr>
<td>1.37/10</td>
<td>1.07/10</td>
<td>0.87/10</td>
<td>0.57/10</td>
<td>0.27/10</td>
</tr>
</tbody>
</table>

**Series II**

- 8-hour LD50 - Rainbow Trout
- 6-hour LD10 and LD50 - Rainbow Trout
- LD10 of Rainbow Trout (percent)

**Series I**

- 8-hour LD50 - Rainbow Trout
- LD50 of Rainbow Trout (percent)
- LD10 and LD50 of Rainbow Trout (percent)

**Series II**

- 8-hour LD50 - Rainbow Trout
- LD50 of Rainbow Trout (percent)
- LD10 and LD50 of Rainbow Trout (percent)

**Series I and II combined with the highest 100% mortality level dropped.**

**Series I only.**

- LD10 and LD50 of Rainbow Trout (percent)

**Series II only.**

- LD10 and LD50 of Rainbow Trout (percent)