

US EPA ARCHIVE DOCUMENT

## VALIDATION SHEET

TEST TYPE: Aquatic Acute Toxicity  
 96-h LC<sub>50</sub>: Bluegill (Lepomis macrochirus)  
 48-h LC<sub>50</sub>: Water flea (Daphnia magna)

CHEMICAL NAME: Vantocil 1B

FORMULATION: 20% aqueous solution

VALIDATOR: Ann Rosenkranz

DATE: September 20, 1979

CITATION: Acute Toxicity of Vantocil 1B, Mix No. 1857, to Bluegill (Lepomis macrochirus) and the Water Flea (Daphnia magna), EG & G, Bionomics, Aquatic Toxicology Laboratory, Wareham, Mass., June, 1977.

VALIDATION CATEGORY: Invalid (bluegill); Supplemental (Daphnia)

RESULTS: Bluegill: 24-h LC<sub>50</sub> = 0.62 mg /l (95% CI = 0.46-0.83 mg/l)  
 48-h LC<sub>50</sub> = 0.62 mg/l (95% CI = 0.46-0.83 mg/l)  
 96-h LC<sub>50</sub> = 0.62 mg/l (95% CI = 0.46-0.83 mg/l)  
 no effect level = 0.24 mg/l

Conc. mg/l	% cumulative mortality		
	24-h	48-h	96-h
1.6	100	100	100
0.75 <sup>1.28</sup>	60	60	60
0.56 <sup>1.02</sup>	30	30	30
0.42 <sup>.82</sup>	10	10	10
0.24 <sup>.66</sup>	0	0	0
0	0	0	0

1. The toxicity of Vantocil 1B to groups of 10 bluegills (mean weight 0.39g) was studied at 22 °C under static conditions using soft quality well water. The test procedures generally complied with the EPA protocol ("Stephan's").
2. The LC<sub>50</sub> values were calculated by converting the test concentrations and corresponding observed percentage mortalities to logs and probits, respectively, and then performing a least squares regression analysis.
3. The chemical is fast-acting since all observed deaths occurred in the first 24 hours.

4. Prior to dying the fish became dark and lethargic and lost equilibrium.

RESULTS: Daphnia: 48-h  $LC_{50}$ : 0.18 mg/l (95% CI = 0.12-0.30 mg/l)  
no effect level = 0.024 mg/l

Conc. mg/l	% Cumulative mortality*	
	24-h	48-h
0.75	100	100
0.32	13	67
0.24	13	47
0.18	33	53
0.14	7	13
0.087	13	13
0.024	0	0
0	0	0

\*Each mortality value represents the average of 3 replicates.

1. The toxicity of Vantocil 1B to Daphnia (<24 hours old) was studied at 22°C under static conditions using soft quality well water. A total of 15 Daphnia were exposed to each concentration, but they were distributed among 3 replicate treatment beakers with 5 Daphnia each. The test procedures generally complied with the EPA protocol ("Stephan's").
2. The  $LC_{50}$  value was calculated by converting the test concentrations and corresponding observed percentage mortalities to logs and probits, respectively, and then performing a least squares regression analyses. The mortality values in the above chart are the mean percentage mortalities of the 3 replicates. Recalculation of the reported 24h- and 48h-  $LC_{50}$  values indicated that the former value is unreliable because the coefficient of determination ( $R^2$ ) was low (.685) indicating there was a poor fit of the data to the straight line. Calculation with the Finney probit method gave a chi-square value of 17.35 (df=5) which indicates that the observed results are significantly different from the expected results.

#### VALIDATION CATEGORY RATIONALE:

The primary reason the bluegill study is classified as invalid is the low dissolved oxygen levels. According to the recommended E.P.A. protocol, Methods for Acute Toxicity Tests with Fish, Macro invertebrates and Amphibians, April, 1975, the dissolved oxygen concentration in each test chamber must be between 60% to 100% saturation during the first 48 hours of the test and between 40% and 100% saturation after 48 hours under static conditions. In this study the dissolved oxygen level ranges from saturation to only 21% of saturation. Additionally there is no indication at which times and concentrations the low D.O. levels occurred. Since the low D.O. levels can also produce stress, the low  $LC_{50}$  value cannot be attributed solely to exposure to Baguacil.

Another problem with the bluegill study is that the formulated product instead of the technical product was used. However, this discrepancy would have caused the study to be classified as supplemental, not invalid. It is for this reason that the Daphnia study is classified as supplemental.

In addition, the mortality data in the Daphnia study should have been reported as the percent mortality for the total number exposed to each concentration, instead of the mean mortality.

Finally, the Daphnia must be in the first instar. It is very likely that the daphnids underwent the first ecdysis if they were nearly 24 hours old. It is preferable to use individuals that are less than 6 hours old to guarantee they are in the first instar (Beltsville Laboratory procedure).

REPAIRABILITY:

The bluegill study cannot be repaired and a new one must be submitted. The mortality data for Daphnia should be recalculated using percent mortality based on 15 individuals per concentration instead of the mean mortality per concentration. However, even with this correction the study will remain supplemental because of the use of the formulated product and the age of the Daphnia.



**EG&G BIONOMICS**

EG&G BIONOMICS, 790 MAIN STREET, WAREHAM, MASSACHUSETTS 02571 • TEL. (617) 295-2550

August 29, 1980

Mr. Allan Wheeler  
Industrial Toxicology  
ICI United States, Inc.  
Wilmington, DE 19897

RE: "Acute Toxicity of Vantocil<sup>R</sup> IB, Mix No. 1857, to Bluegill  
(Lepomis macrochirus) and the Water Flea (Daphnia magna),"  
June 1977.

Dear Mr. Wheeler:

This letter is in response to our phone conversation on 25 August 1980 concerning the lack of increased mortality of bluegill beyond 24-hours exposure to Vantocil. A response such as this is not uncommon in acute toxicity tests and it is generally the result of one of two factors:

1. Persistence of the toxic component of the test material. Cumulative mortalities of fish may not be observed if the chemical nature of the toxic component of the test material is such that shortly after it is dissolved in water the toxic properties are lost due to such factors

not readily absorbed or metabolized by the fish during the initial 96 hours of exposure.

I assure you that the results of the acute toxicity tests are reliable and that the response observed is by no means unique to Vantocil.

If I can be of any further assistance, please do not hesitate to contact me.

Sincerely,

Gerald A. LeBlanc  
Aquatic Toxicologist

GAL:jeb

## STUDY VALIDATION

DATA REVIEW NUMBER: ES-H-1

TEST: Aquatic invertebrate 48-hr LC<sub>50</sub>

SPECIES: Daphnia magna

## RESULTS:

48 hr. LC<sub>50</sub> = 0.18 mg/l  
95% conf. limits = 0.12 - 0.30 mg/l

CHEMICAL: Vantocil 1B (20% a.i.)

TITLE: Acute toxicity of Vantocil 1B, mix No. 1857,  
to bluegill ( Lepomis Macrochirus ) and the  
water flea ( Daphnia magna ).

ACCESSION NO: 234289, Report # C-23

STUDY DATE: June, 1977

RESEARCHER: G. A. LeBlanc and B. H. Sleight, III; EG&G  
Bionomics, Aquatic Toxicology Laboratory.

REGISTRANT: ICI Americas, Inc.

VALIDATION CATEGORY: Supplemental

CATEGORY REPAIRABILITY: Yes, to Core

## ABSTRACT:

Test protocol closely followed "Stephan" and is acceptable. Test parameters reported such as pH, DO, water hardness, temperature, etc. were maintained within acceptable limits.

Statistical analysis of the results (attached) closely resemble the results reported.

NOTE: The material used was the formulated product. Thus, upgrading this study to core should depend on the need for this particular study and clarification of the statistical analysis.

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LW H  
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0.219

0.179

0.268

LD50  
LDCL  
UPCL

0.093

0.065

0.135

LD10  
LDCL  
UPCL

0.515

0.342

0.776

LD  
LD  
UP

42

## STUDY VALIDATION

DATA REVIEW NUMBER: ES-F-1

TEST: Fish acute 96-hr LC<sub>50</sub>

SPECIES: Bluegill ( Lepomis macrochirus )

RESULTS: 96-hr LC<sub>50</sub> = 0.62 mg/l  
95% Conf. limites = 0.46 - 0.83 mg/l

CHEMICAL:

Vantocil 1B (20% a.i.)

TITLE: Acute toxicity of Vantocil 1B, Mix No. 1857, to bluegill (Lepomis macrochirus) and the water flea ( Daphnia magna ).

ACCESSION NO.: 234289, Report # C-23

STUDY DATE: June, 1977

RESEARCHER: R. J. Buccafusco and Bevier Hasbrouck Sleight, III;  
EG&G, Bionomics, Aquatic Toxicology Laboratory.

REGISTRANT: ICI Americas Inc.

VALIDATION CATEGORY: Supplemental

CATEGORY REPAIRABILITY: Yes, to core

Test protocol used closely followed "Stephan" and is acceptable. Test parameters reported such as pH, DO, water hardness, temperature etc. were maintained within acceptable limits.

NOTE: Statistical analysis of the results (attached) closely resemble the results reported.

The material used was the formulated product; thus, upgrading this study to core should depend on the need for this particular study and clarification of the statistical analysis.

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6.715  
 6.162 YINT  
 1.409 LD  
 0.127 CHIS  
  
 0.67 LD50  
 0.56 LDCL  
 0.80 UPCL  
  
 0.43 LD10  
 0.33 LD5  
 0.562 UPCL  
  
 1.042  
 0.726  
 1.496

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