UNITED STATES ENVIRONMENTAL PROTECTION AGENCY
WASHINGTON, D.C. 20460

AUG 16 1999

OFFICE OF
PREVENTION, PESTICIDES AND
TOXIC SUBSTANCES

SUBJECT: **ECOLOGICAL REVIEW OF: 072560 Silver Sodium Hydrogen Zirconium Phosphate (AlphaSan™ RC 5000 and AlphaSan™ RC 7000)**

DP Bar-code: D253522 **Reg. No. Or File Symbol: 011631-E & 011631-U**

Manufacturing-Use - [X] **OR End-Use Product - [ ]**

TO: Tony Kish/Marshall Swindell
Regulatory Branch I, PM Team No. - 33
Antimicrobials Division (7510W)

FROM: Wanda Jakob, Biologist Wanda Jakob 08/09/99
Risk Assessment and Science Support Branch, Team 2
Antimicrobials Division (7510W)

THRU: Tim McMahon, Acting Team Leader 8/09/99
Risk Assessment and Science Support Branch, Team 2
Antimicrobials Division (7510W)

Norm Cook, Chief 08.16.99
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SUMMARY OF INFORMATION REVIEWED AND FINDINGS

I. Introduction

Milliken Chemical has submitted an application for registration of two new pesticide active ingredients, Silver Sodium Hydrogen Zirconium Phosphate (3.8% silver by weight and 10% silver by weight). The trade names are AphaSan™ RC 5000 and AlphaSan™ RC 7000 respectfully. The proposed products are antimicrobial powders designed to be incorporated into materials (i.e., plastic, fibers, coatings, adhesives and sealants) during the manufacturing process to impart antimicrobial activity to the manufactured products. AlphSan™ RC 5000 and 7000 are used to suppress the growth of bacteria, algae, fungus, mold and mildew. The Regulatory Branch

I/Antimicrobials Division has requested the Risk Assessment and Science Support Branch/Antimicrobials Division to complete an ecological review of the proposed registration application.

II. Summary of Information Reviewed and Findings

♦ AlphSan™ RC 5000 and 7000 are characterized as an Indoor Materials Preservative. There are four required ecological studies listed by the Agency for materials preservative pesticides. The studies are listed as follows:

1. Avian oral LD50, bobwhite quail or mallard (*Colinus virginianus* or *Anas platyrhynchos*), Guideline 71-1/850.2100
2. Freshwater fish LC50, rainbow trout (*Oncorhynchus mykiss*), Guideline 72- 1/ 850.1750
3. Freshwater fish LC50, bluegill sunfish (*Lepomis macrochirus*), Guideline 72- 1/ 850.1750
4. Acute EC50, freshwater invertebrate (*Daphnia magna*), Guideline 72-2/850.1010

♦ The following table summarizes the data and the Agency's classification for each of the studies submitted by Milliken Chemical for the active ingredient, Silver Sodium Hydrogen Zirconium Phosphate.

Summary of Data for Silver Sodium Hydrogen Zirconium Phosphate (AlphaSan™ RC 5000 and 7000)

Guideline	MRID Number	Test Species	Test Material (% a.i.)	Test Type	Test Results	Toxicity Level	Study Status	Citation
71-1/ 850.2100	445829-21	Bobwhite Quail	100	Acute Oral	LD50: >2000 ppm NOEL: 2000 ppm	Practically Nontoxic	**Supplemental**	Helsten, BR; 1998
72-1/ 850.1750	445829-18	Rainbow Trout	≥99	Fish Acute	LC50: 1.2 ppm NOEC: 1.0 ppm	Moderately Toxic	**Invalid**	Bell, G; Groom, S; Smith, B; 1995
72-1/ 850.1750	445829-19	Bluegill Sunfish	100 (assumed)	Fish Acute	LC50: 1.7 ppm NOEC: 1.0 ppm	Moderately Toxic	**Invalid**	Bell, G; Groom, S; Smith, B; 1996
72-2/ 850.1010	445829-20	*Daphnia magna*	≥99	Invertebrate Acute	LC50: 0.13 ppm NOEC: N/R*	Highly Toxic	**Invalid**	Bell, G; Groom, S; Smith, B; 1995

* N/R = Not Reported

Citations

Bell, G.; Groom, S; Smith, B. July 1995. Application for Pesticide Registration: *AlphaSan*™ RC 5000, EPA File Symbol 11631-, Volume 20: Fish Toxicity Test. Huntingdon Research Center, Ltd., P.O. Box 2, Huntingdon, Cambridgeshire, PE18 6ES, England. Laboratory Report Number: TSI 64(c)/950717. MRID 445829-18.

Bell, G.; Groom, S; Smith, B. July 1995. Application for Pesticide Registration: *AlphaSan*™ RC 5000, EPA File Symbol 11631-, Volume 21: Fish Toxicity Test. Huntingdon Research Center, Ltd., P.O. Box 2, Huntingdon, Cambridgeshire, PE18 6ES, England. Laboratory Report Number: TSI 64(d)/941226. MRID 445829-19.

Bell, G.; Groom, S; Smith, B. July 1995. Application for Pesticide Registration: *AlphaSan*™ RC 5000, EPA File Symbol 11631-, Volume 22: Acute Toxicity to *Daphnia Magna*. Huntingdon Research Center, Ltd., P.O. Box 2, Huntingdon, Cambridgeshire, PE18 6ES, England. Laboratory Report Number: TSI 64(b)/950718. MRID 445829-20.

Helsten, BR. March 1998. Avian Acute Oral Toxicity Test in Bobwhite Quail with Experimental Additive 9823-37. Bio-Life® Associates, Ltd. (BLAL), N6230 County Road G, Neillsville, WI 54456-8773. Laboratory Report Number: BLAL No. 163-001-03. MRID 445829-21.

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III. Conclusion/Recommendations

It was the Agency's understanding that the product to be tested would be AlphaSan™ RC 2000 (10% silver by weight). The toxicity data to be submitted would be used to evaluate the pending registration applications for AlphaSan™ RC 2000 (10% silver by weight), AlphaSan™ RC 5000 (3.8% silver by weight), and AlphaSan™ RC 7000 (10% silver by weight plus the presence of [redacted].

1. The avian oral LD_{50} was conducted on the Bobwhite quail. This study is scientifically sound, however, it has been classified as **Supplemental**. The rationale for the Supplemental classification is based on the fact that the authors did not document the identity of the test substance or whether it is equivalent to AlphaSan™ RC 2000.
Recommendation: provide information on the chemical composition of the test substance from Milliken Chemical Associates, Ltd. The substance tested should be Silver Sodium Hydrogen Zirconium Phosphate (AlphaSan™ RC 2000, 10% silver by weight).

2. The study for the freshwater fish LC_{50} conducted with rainbow trout was not scientifically sound and was classified as **Invalid**. The test substance (Novaron AG300) did not contain silver. Silver is one of the constituents of the substance being submitted for registration. Furthermore, analysis of water samples indicated measured concentrations that were as low as 61% of nominal in fresh solutions and as low as 15% of nominal in old solutions (Agency guidelines indicate that test concentrations must remain at least 80% of nominal throughout the test). Analysis of the dilution water, which was dechlorinated tap water, indicated higher than acceptable levels of several elements (Agency guidelines state that dilution water should not be dechlorinated tap water). Additionally, a range-finding test was not reported and analytical percent recovery of the test material was not reported.
Recommendation: repeat the Freshwater fish LC50, rainbow trout (*Oncorhynchus mykiss*) study and test the active ingredient, Silver Sodium Hydrogen Zirconium Phosphate (AlphaSan™ RC 2000, 10% silver by weight).

3. The study for the freshwater fish LC_{50} conducted with bluegill sunfish was not scientifically sound and was classified as **Invalid**. The test substance (Novaron AG300) did not contain silver. Silver is one of the constituents of the substance being submitted for registration. Furthermore, analysis of water samples indicated measured concentrations that were as low as 53% of nominal in fresh solutions and as low as 30% of nominal in old solutions (Agency guidelines indicate that test concentrations must remain at least 80% of nominal throughout the test). Range-finding tests and recovery of the chemical were not reported. The turbidimetric method of measuring test substance concentrations might not be valid for solutions more than an hour old. Analysis of the dilution water, which was dechlorinated tap water, indicated higher than acceptable levels of several elements (Agency guidelines state that dilution water should not be dechlorinated tap water). Additionally, a rang-finding test was not reported and analytical percent recovery of the test material was not reported.
Recommendation: repeat the Freshwater fish LC50, bluegill sunfish (*Lepomis macrochirus*)

study and test the active ingredient, Silver Sodium Hydrogen Zirconium Phosphate (AlphaSan™ RC 2000, 10% silver by weight).

4. The Acute EC50, freshwater invertebrate (*Daphnia magna*) study was not scientifically sound and was classified as **Invalid**. The test substance (Novaron AG300) did not contain silver. Silver is one of the constituents of the substance being submitted for registration. Furthermore, measured concentrations ranged form 87% to 127% of nominal at 0 hr. But were only 0% to 15% of nominal at 48 hr. Additionally, the authors did not report all of the required acclimation information, including the length of the acclimation period, pretest mortality, health of the test organisms, or the presence of ephippia. The hardness of the dilution water was not reported, a range-finding test was not conducted or data was not reported, the percent recovery of the chemical was not reported, and a NOEC was not reported despite adequate data to determine the NOEC for immobilization.
Recommendation: repeat the Acute EC50, freshwater invertebrate (*Daphnia magna*) study and test the active ingredient, Silver Sodium Hydrogen Zirconium Phosphate (AlphaSan™ RC 2000, 10% silver by weight).

cc: RASSB Chemical File (072560)
Tim McMahon/RASSB

DATA EVALUATION REPORT

ZIRCONIUM PHOSPHATE
(NOVARON AG300)

Study Type: Acute Coldwater Fish LC_{50} Test (*Oncorhynchus mykiss*)

Prepared for

Antimicrobial Division
Office of Pesticide Programs
U.S. Environmental Protection Agency
1921 Jefferson Davis Highway
Arlington, VA 22202

Prepared by

ICF Consulting Group
9300 Lee Highway
Fairfax, VA 22031

Principal Reviewer	Matthew C. Preisser *Matthew Preisser, M.E.M.*	Date 3/31/99
Independent Reviewer	Margaret McVey *Margaret McVey, Ph.D.*	Date 3/31/99
Project Manager (QA/QC Manager)	Ellen Mantus *Ellen Mantus, Ph.D.*	Date 4/5/99

Contract Number: 68-W6-0022
Work Assignment No.: 3-17
EPA Project Manager: Peter Thompson

Disclaimer

This review may have been changed following contractor's submission to the Antimicrobial Division of the Office of Pesticide Programs.

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DP Barcode: D253522

MRID No.: 445829-18

DATA EVALUATION RECORD
§ 72-1(C) -- ACUTE LC_{50} TEST WITH A COLDWATER FISH

1. CHEMICAL: Zirconium phosphate[1] PC Code No.:

2. TEST MATERIAL: NOVARON AG300 (Batch No. 7130519; white powder) Purity: ≥ 99%

3. CITATION

Authors[2]:	G. Bell, S. Groom, and B. Smith
Title:	Application for Pesticide Registration: *AlphaSan™ RC 5000*, EPA File Symbol 11631- , Volume 20: Fish Toxicity Test
Study Completion Date:	July 13, 1995
Laboratory:	Huntingdon Research Centre, Ltd., P.O. Box 2, Huntingdon, Cambridgeshire, PE18 6ES, England
Sponsor:	Milliken Chemical, A Division of Milliken & Company, P.O. Box 1927, Spartanburg, SC 29304-1927
Laboratory Report ID:	TSI 64(c)/950717
MRID No.:	445829-18
DP Barcode:	D253522

4. REVIEWED BY: W. Jakob, Antimicrobials Division (7510C)

Signature: Wanda J Jakob Date: 08-09-99

5. APPROVED BY: P. Thompson, Ph.D., Antimicrobials Division (7510C)

Signature: Wanda J Jakob Date: 08-09-99

6. STUDY PARAMETERS

Scientific Name of Test Organism:	*Oncorhynchus mykiss*
Age or Size of Test Organism:	Mean weight: 0.69 g; mean length: 3.8 mm
Definitive Test Duration:	96 hr (03/13/95 to 03/17/95)
Study Method:	Static renewal
Type of Concentrations:	Nominal

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[1] The acute toxicity test report is being submitted in support of the reregistration of AlphaSan™ RC 5000, which contains silver sodium hydrogen zirconium phosphate (3.8% silver by weight). The toxicity test report under review indicates that the test substance was NOVARON AG300, which is ≥ 99% "partially ion-exchange" zirconium phosphate. There is no evidence that the test substance contained any silver, which is the true active ingredient.

[2] The authors were the personnel responsible for conducting the experiments and were affiliated with the performing laboratory, Huntingdon Research Centre, Ltd. The MRID report was prepared by Keller and Heckman LLP, 1001 G Street, N.W., Suite 500 West, Washington, D.C., 20001.

7. CONCLUSIONS

Results Synopsis[3]:
LC_{50}: 1.2 ppm 95% C.I.: 1.1 - 1.4 ppm
NOEC: 1.0 ppm Probit Slope: N/A

Validated Results Synopsis:
LC_{50}: 0.62 ppm 95% C.I.: Not reliable
NOEC: ≤ 0.53 ppm Probit Slope: N/A

8. ADEQUACY OF THE STUDY

A. Classification: Invalid

B. Rationale: According to information provided on page 10 of the report, the test substance did not contain silver, which is one of the constituents of the substance being submitted for reregistration. Furthermore, analysis of water samples indicated measured concentrations that were as low as 61% of nominal in fresh solutions and as low as 15% of nominal in old solutions. Analysis of the dilution water, which was dechlorinated tap water, indicated higher than acceptable levels of several elements.

C. Repairability: Request from sponsor information on whether the test substance supplied to the test laboratory contained silver and if so, how much silver was in the test substance.

9. GUIDELINE DEVIATIONS

1. Contrary to what is recommended in these guidelines (OPP § 72-1(c)) and in the EPA 1996 harmonized ecological effects guidelines (OPPTS 850.1075), the source of the dilution water was dechlorinated tap water.

2. The analytic method for estimating the concentration of zirconium phosphate in solution was not adequate -- see Reviewer's Comments (Section 14).

3. The OPPTS 850.1075 guidelines establish dilution water concentration limits for several elements. Analysis of the municipal water supply (the same source as the dilution water), presented in Appendix 3 of the report, indicated that the dilution water exceeded the maximum limits for several elements, including iron, copper, arsenic, cobalt, nickel, lead, zinc, aluminum, and iron. Additionally, the total organic carbon was above the maximum limit.

4. The test substance was not measured in each replicate before and after each renewal, as is recommended for static-renewal tests in the OPPTS 850.1075 guidelines. This was done only for the first 24-hr period due to high mortality at all but the lowest concentration level after 24 hr. Furthermore, the OPPTS 850.1075 guidelines indicate that test concentrations must remain at least 80% of nominal throughout the test. Measured concentrations ranged as low as 61% of nominal in samples of fresh solution and 15% of nominal in samples of old solution. See Reported Results (Section 12) and Reviewer's Comments (Section 14).

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[3] Study results reported in mg/L and converted to ppm by the technical reviewer.

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5. A range-finding test was not reported.

6. Analytical percent recovery of the test material was not reported.

10. SUBMISSION PURPOSE: Reregistration

11. MATERIALS AND METHODS

A. Test Organisms

Guideline Criteria	Reported Information
Species Preferred species is the rainbow trout (*Oncorhynchus mykiss*)	Rainbow trout (*Oncorhynchus mykiss*)
Mean Weight 0.5-5 g	Mean weight: 0.69 ± 0.16 (SD) g (measurements of control fish at the end of the 4-day test period)
Mean Standard Length Longest not > 2x shortest	Mean standard length: 3.8 ± 0.34 (SD) mm (measurements of control fish at the end of the test period) Longest not > 2x shortest
Supplier	Westacre Trout Farm, Norfolk, U.K.
All fish from same source?	Yes
All fish from the same year class?	Yes

B. Source/Acclimation

Guideline Criteria	Reported Information
Acclimation Period Minimum 14 days	14 days
Wild caught organisms were quarantined for 7 days?	N/A
Were there signs of disease or injury?	No
If treated for disease, was there no sign of the disease remaining during the 48 hr prior to testing?	N/A
Feeding No feeding during the study	No feeding 24 hr prior to test or during the study

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Guideline Criteria	Reported Information
Pretest Mortality < 3% mortality 48 hr prior to testing	0%

C. Test System

Guideline Criteria	Reported Information
Source of dilution water Soft reconstituted water or water from a natural source, *not* dechlorinated tap water	Laboratory tap water filtered, dechlorinated, and softened by passage through an Elga® water purification system (water quality analysis, prior to filtration, dechlorination, and softening, is given in Appendix 3 of the report)
Does water support test animals without observable signs of stress?	Not reported, but no signs of toxicity observed in control fish
Water Temperature 12°C	12 - 13 °C
pH Prefer 7.2 to 7.6	pH range = 7.2 to 8.0
Dissolved Oxygen Static: ≥ 60% during 1st 48 hr and ≥ 40% during 2nd 48 hr, flow-through: ≥ 60%	DO range = 8.8 to 10.4 mg/L (approximately 83% to 98% saturation at sea level and 13 °C)
Total Hardness Prefer 40 to 48 mg/L as $CaCO_3$	168 ± 7 mg $CaCO_3$/L (Note: EPA's 1996 harmonized guidelines recommend a total hardness range of 40 to 180 mg/L as $CaCO_3$)
Test Aquaria 1. Material: glass or stainless steel 2. Size: volume of 18.9 L (5 gal) or 30 x 60 x 30 cm 3. Fill volume: 15-30 L of solution	1. Material: Glass 2. Size: 25 x 46 x 25 cm 3. Fill volume: 20 L
Type of Dilution System Must provide reproducible supply of toxicant	N/A
Flow Rate Consistent flow rate of 5-10 vol/24 hr, meter systems calibrated before study and checked twice daily during test period	N/A
Biomass Loading Rate Static: ≤ 0.8 g/L at ≤ 17°C, ≤ 0.5 g/L at > 17°C; flow-through: ≤ 1 g/L/day	0.35 g/L

Guideline Criteria	Reported Information
Photoperiod 16 hr light, 8 hr dark	16 hr light, 8 hr dark
Solvents Not to exceed 0.5 ml/L for static tests or 0.1 ml/L for flow-through tests	Solvent: None Maximum concentration: N/A

D. Test Design

Guideline Criteria	Reported Information
Range Finding Test If LC_{50} >100 mg/L with 30 fish, then no definitive test is required.	Not reported
Nominal Concentrations of Definitive Test Control & 5 treatment levels; dosage should be 60% of the next highest concentration; concentrations should be in a geo-metric series	Negative control and 5 nominal test concentrations (ppm): 1.0; 1.8; 3.2; 5.6; and 10.0; each concentration is 56% of the next higher concentration; concentrations were in a geometric series
Number of Test Organisms Minimum 10/level, may be divided among containers	20/level (10 test organisms per test vessel and 2 test vessels per level)
Test organisms randomly or impartially assigned to test vessels?	Yes
Biological observations made every 24 hr?	Yes
Water Parameter Measurements 1. Temperature: Measured constantly or, if water baths are used, every 6 hrs, may not vary > 1°C 2. DO and pH: Measured at beginning of test and ever 48 hr in the high, medium, and low doses and in the control	1. Temperature was measured in all replicates and reported every 24 hr for vessels with surviving fish (Appendix 1). It did not vary by more than 1°C. 2. DO and pH were measured in all replicates and reported every 24 hr for vessels with surviving fish (Appendix 1).
Chemical Analysis Needed if solutions were aerated, if chemical was volatile, insoluble, or known to absorb, if precipitate formed, if containers were not steel or glass, or if flow-through system was used	Yes. Water samples were taken in duplicate from the control and each exposure level (samples from replicate vessels were pooled) at 0 and 72 hr (fresh media) and at 24 and 96 hr (expired media). No aeration was provided.

12. REPORTED RESULTS

A. General Results

Guideline Criteria	Reported Information
Quality assurance and GLP compliance statements were included in the report?	Yes
Recovery of Chemical	Not reported
Control Mortality Not more than 10% control organisms may die or show abnormal behavior.	0%
Raw data included?	Yes
Signs of toxicity (if any) were described?	Yes

Mortality[4]

Concentration (ppm)		Number of Fish	Cumulative Number Dead			
			Hour of Study			
Nominal	Measured (0 hr - 24 hr)		24	48	72	96
Control	not detected	20	0	0	0	0
Solvent Control	--	N/A	N/A	N/A	N/A	N/A
1.0	0.61 - 0.46	20	0	0	1	2
1.8	0.85 - 0.65	20	6	20	20	20
3.2	2.0 - 1.0	20	14	20	20	20
5.6	3.7 - 1.6	20	20	20	20	20
10.0	7.4 - 1.4	20	20	20	20	20

Other significant results: Fish exhibited non-lethal effects after 6 hours, including lying on the bottom of the vessels, swimming at the surface, increased pigmentation, excessive mucus production, and lethargy.

[4] All concentrations were reported in mg/L, which the technical reviewer converted to the ppm equivalents. The test media were renewed each day and measured concentrations (for all 5 test levels) were estimated only at the beginning and end of the first 24-hr period (see Reviewer's Comments section and Appendix 2 of the report).

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B. Statistical Results[5]

Method: LC_{50}, logistic model (Berkson, 1944[6]); 95% C.I., likelihood ratio method (Williams, 1986[6]); NOEL, the highest concentration tested at which no significant mortality or sublethal adverse effects were observed (an incidence rate of more than one affected fish out of ten, in any replicate, was considered significant).

96-hr LC_{50}: 1.2 ppm (nominal) 95% C.I.: 1.1 - 1.4 ppm (nominal)
Probit Slope: N/A NOEC: 1.0 ppm

Other results (nominal):
3-hr LC_{50} (95% C.I.): > 10 (N/A) ppm
6-hr LC_{50} (95% C.I.): > 10 (N/A) ppm
24-hr LC_{50} (95% C.I.): 2.4 (2.0 - 2.8) ppm
48-hr LC_{50} (95% C.I.): 1.3 (1.2- 1.6) ppm
72-hr LC_{50} (95% C.I.): 1.2 (1.1 - 1.4) ppm

13. VERIFICATION OF STATISTICAL RESULTS

Parameter	Results (measured)
Binomial Test LC_{50} (C.I.)	Not used
Moving Average Angle LC_{50} (95% C.I.)	Not used
Probit LC_{50} (95% C.I.)	Not appropriate
Probit Slope	Not appropriate
Trimmed Spearman-Karber LC_{50} (95% C.I.)	0.62 ppm (95% C.I. not reliable)
NOEC	≤ 0.53 ppm

Comments: To verify the statistical results, we used the mean of the 0-hr and 24-hr concentration measurements for each test level. However, as described below in Section 14, we do not have confidence in the measured concentrations. Ten percent trim was used in the Trimmed Spearman-Karber analysis.

14. REVIEWER'S COMMENTS

All results reported by the authors are expressed in terms of nominal concentrations. Chemical analysis of fresh media (samples taken after media renewal at 0 and 72 hr) and expired media (samples taken from the same "used" media at 24 and 96 hr) revealed that the measured concentrations ranged from 61% to 111%

[5] All concentrations were reported in mg/L, which the technical reviewer converted to the ppm equivalents

[6] Berkson, J. 1944. Application of the logistic function to bio-assay. *J. Amer. Statist. Assoc.* 39: 357-365.

Williams, D.A. 1986. Interval estimation of the median lethal dose. *Biometrics* 42:641-645.

of nominal for the fresh media and 15% to 78% for the expired media. The authors noted that while near nominal concentrations were achieved for the fresh media, difficulties in maintaining homogeneity and stability of NOVARON accounted for the losses in the expired media.

The Huntingdon Research Centre analytical laboratory used a turbidimetric method to measure NOVARON in samples of water from the test vessels. The laboratory considered the method adequate because NOVARON forms a homogenous suspension when added to water. Each sample was ultrasonicated for five minutes and well shaken prior to analysis. No data were presented, however, to indicate that the zirconium phosphate does not become hydrated and "dissolve" in the water over time such that a turbidimetric method of analysis would not detect it. Given the "all or nothing" type of response seen in this test, the uncertainty in the measured concentrations is unacceptably high.

We also do not understand how zirconium phosphate alone could be used to assess the acute aquatic toxicity of AlphaSan™ RC 5000, which contains 3.8% silver by weight. Silver is extremely toxic to aquatic organisms. The proposed freshwater acute ambient water quality criterion for silver is 0.92 μg/L (ppt).

DATA EVALUATION REPORT

ZIRCONIUM PHOSPHATE
(NOVARON AG300)

Study Type: Acute Warmwater Fish LC_{50} Test (*Lepomis macrochirus*)

Prepared for

Antimicrobial Division
Office of Pesticide Programs
U.S. Environmental Protection Agency
1921 Jefferson Davis Highway
Arlington, VA 22202

Prepared by

ICF Consulting Group
9300 Lee Highway
Fairfax, VA 22031

Principal Reviewer	Matthew C. Preisser *Matthew Preisser, M.E.M.*	Date 3/31/99
Independent Reviewer	Margaret McVey *Margaret McVey, Ph.D.*	Date 3/31/99
Project Manager (QA/QC Manager)	Ellen Mantus *Ellen Mantus, Ph.D.*	Date 4/5/99

Contract Number: 68-W6-0022
Work Assignment No.: 3-17
EPA Project Manager: Peter Thompson

Disclaimer

This review may have been changed following contractor's submission to the Antimicrobial Division of the Office of Pesticide Programs.

DP Barcode: D253522

MRID No.: 445829-19

DATA EVALUATION RECORD
§ 72-1(A) -- ACUTE LC_{50} TEST WITH A WARMWATER FISH

1. CHEMICAL: Zirconium phosphate[1]

PC Code No.:

2. TEST MATERIAL: NOVARON AG300
(Batch No. 7160203; white powder)

Purity: 100% (assumed)

3. CITATION

Authors[2]:	G. Bell, S. Groom, and B. Smith
Title:	Application for Pesticide Registration: *AlphaSan™ RC 5000*, EPA File Symbol 11631- , Volume 21: Fish Toxicity Test
Study Completion Date:	August 4, 1996
Laboratory:	Huntingdon Research Centre, Ltd., P.O. Box 2, Huntingdon, Cambridgeshire, PE18 6ES, England
Sponsor:	Milliken Chemical, A Division of Milliken & Company, P.O. Box 1927, Spartanburg, SC 29304-1927
Laboratory Report ID:	TSI 64(d)/941226
MRID No.:	445829-19
DP Barcode:	D253522

4. REVIEWED BY: W. Jakob, Antimicrobials Division (7510C)

Signature: Wanda J Jakob Date: 08/09/99

5. APPROVED BY: P. Thompson, Ph.D., Antimicrobials Division (7510C)

Signature: Wanda J Jakob Date: 08/09/99

6. STUDY PARAMETERS

Scientific Name of Test Organism:	*Lepomis macrochirus*
Age or Size of Test Organism:	Mean weight: 0.64 g; mean length: 3.1 mm
Definitive Test Duration:	96 hr (07/30/96 to 08/04/96)
Study Method:	Static renewal
Type of Concentrations:	Nominal

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[1] The acute toxicity test report is being submitted in support of the reregistration of AlphaSan™ RC 5000, which contains silver sodium hydrogen zirconium phosphate (3.8% silver by weight). The toxicity test report under review indicates that the test substance was NOVARON AG300, which was assumed by the authors to be 100% "partially ion-exchange" zirconium phosphate. There is no evidence that the test substance contained any silver, which is the true active ingredient.

[2] The authors were the personnel responsible for conducting the experiments and were affiliated with the performing laboratory, Huntingdon Research Centre, Ltd. The MRID report was prepared by Keller and Heckman LLP, 1001 G Street, N.W., Suite 500 West, Washington, D.C., 20001.

7. CONCLUSIONS

Results Synopsis[3]:
96-hr LC_{50}: 1.7 ppm 95% C.I.: 1.5 - 1.8 ppm
Probit Slope: N/A NOEC: 1.0 ppm

Validated Results Synopsis:
96-hr LC_{50}: 0.87 ppm 95% C.I.: 0.79 - 0.96 ppm
Probit Slope: N/A NOEC (mortality): 0.58 ppm

8. ADEQUACY OF THE STUDY

A. Classification: Invalid

B. Rationale: According to information provided on page 9 of the report, the test substance did not contain silver, which is one of the constituents of the substance being submitted for reregistration. Furthermore, analysis of water samples indicated measured concentrations that were as low as 53% of nominal in fresh solutions and as low as 30% of nominal in old solutions. Range-finding tests and recovery of the chemical were not reported. The turbidimetric method of measuring test substance concentrations might not be valid for solutions more than an hour old. Analysis of the dilution water, which was dechlorinated tap water, indicated higher than acceptable levels of several elements.

C. Repairability: Request from sponsor information on whether the test substance supplied to the test laboratory contained silver and if so, how much silver was in the test substance.

9. GUIDELINE DEVIATIONS

1. Contrary to what is recommended in these guidelines (OPP § 72-1(a)) and in the EPA 1996 harmonized ecological effects guidelines (OPPTS 850.1075), the source of the dilution water was dechlorinated tap water.

2. The test substance was not measured in each replicate before and after each renewal, as is recommended for static-renewal tests in the OPPTS 850.1075 guidelines. Although it is understandable that this was not done at the high test concentrations, due to high (100%) mortality 24 hr after test initiation, measurements were not conducted for each renewal period at the lowest two concentrations (e.g., for the second 24-hr period (i.e., 24 hr to 48 hr)). Furthermore, the OPPTS 850.1075 guidelines indicate that test concentrations must remain at least 80% of nominal throughout the test. Measured concentrations for samples of fresh media ranged as low as 53% of nominal, while samples of 24-hr old media ranged as low as 30% of nominal. See Reviewer's Comments (Section 14).

3. The water temperature (19 - 20 °C) was slightly below the OPPTS 850.1075 recommended temperature for bluegill sunfish (22 ± 2 °C)

4. A range-finding test was not reported.

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[3] Study results reported in mg/L and converted to ppm by the technical reviewer.

5. Analytical percent recovery of the test material was not reported.

6. The OPPTS 850.1075 guidelines establish dilution water concentration limits for several elements. Analysis of the municipal water supply (the same source as the dilution water), presented in Appendix 3 of the report, indicated that the quantification limits of the chemical analysis methods used were higher than the OPPTS recommended limits. Thus, it cannot be determined whether many of the analyses exceeded those limits for residual chlorine, ammonia, aluminum, chromium, copper, iron, lead, nickel, zinc, cadmium, and mercury. Additionally, the total organophosphorus pesticide level might have been above the maximum limit.

10. SUBMISSION PURPOSE: Reregistration

11. MATERIALS AND METHODS

A. Test Organisms

Guideline Criteria	Reported Information
Species Preferred species is the bluegill sunfish (*Lepomis macrochirus*)	Bluegill sunfish (*Lepomis macrochirus*)
Mean Weight 0.5-5 g	Mean weight: 0.64 ± 0.21 (SD) g (measurements of control fish at the end of the test period)
Mean Standard Length Longest not > 2x shortest	Mean standard length: 3.1 ± 0.30 (SD) mm (measurements of control fish at the end of the test period) Longest not > 2x shortest
Supplier	Charles River, UK, Ltd.
All fish from same source?	Yes
All fish from the same year class?	Yes

B. Source/Acclimation

Guideline Criteria	Reported Information
Acclimation Period Minimum 14 days	13 days
Wild caught organisms were quarantined for 7 days?	N/A
Were there signs of disease or injury?	No

Guideline Criteria	Reported Information
If treated for disease, was there no sign of the disease remaining during the 48 hours prior to testing?	N/A
Feeding No feeding during the study	No feeding 24 hr prior to test or during the study
Pretest Mortality No more than 3% mortality 48 hr prior to testing	1.5% in the 7 days prior to the test

C. Test System

Guideline Criteria	Reported Information
Source of dilution water Soft reconstituted water or water from a natural source, **not** dechlorinated tap water	Laboratory tap water filtered, dechlorinated, and softened by passage through an Elga® water purification system (water quality analysis, prior to filtration, dechlorination, and softening, is given in Appendix 3 of the report)
Does water support test animals without observable signs of stress?	Not reported, but no signs of toxicity observed in control fish
Water Temperature 17°C or 22°C	Range = 19 to 20 °C
pH Prefer 7.2 to 7.6	Range = 7.4 to 8.0
Dissolved Oxygen Static: ≥ 60% during 1st 48 hr and ≥ 40% during 2nd 48 hr, flow-through: ≥ 60%	Range: 6.4 to 8.4 mg/L (approximately 70% to 93% saturation at sea level and 20 °C)
Total Hardness Prefer 40 to 48 mg/L as $CaCO_3$	132 ± 11 (SD) mg/L as $CaCO_3$ (Note: The OPPTS 850.1075 guidelines recommend a total hardness range of 40 to 180 mg/L as $CaCO_3$)
Test Aquaria 1. Material: glass or stainless steel 2. Size: volume of 19 L (5 gal) or 30 x 60 x 30 cm 3. Fill volume: 15-30 L of solution	1. Material: Glass 2. Size: 25 x 46 x 25 cm 3. Fill volume: 20 L
Type of Dilution System Must provide reproducible supply of toxicant	N/A

Guideline Criteria	Reported Information
Flow Rate Consistent flow rate of 5-10 vol/24 hr, meter systems calibrated before study and checked twice daily during test period	N/A
Biomass Loading Rate Static: ≤ 0.8 g/L at ≤ 17°C, ≤ 0.5 g/L at > 17°C; flow-through: ≤ 1 g/L/day	0.32 g/L
Photoperiod 16 hr light, 8 hr dark	16 hr light, 8 hr dark
Solvents Not to exceed 0.5 ml/L for static tests or 0.1 ml/L for flow-through tests	Solvent: None Maximum concentration: N/A

D. Test Design

Guideline Criteria	Reported Information
Range Finding Test If LC_{50} >100 mg/L with 30 fish, then no definitive test is required.	Not reported
Nominal Concentrations of Definitive Test Control & 5 treatment levels; dosage should be 60% of the next highest concentration; concentrations should be in a geo-metric series	Negative control and 5 nominal test concentrations (ppm): 1.0; 1.8; 3.2; 5.6; and 10.0; each concentration is 56% of the next higher concentration; concentrations were in a geometric series
Number of Test Organisms Minimum 10/level, may be divided among containers	20/level (10 test organisms per test vessel and 2 test vessels per level)
Test organisms randomly or impartially assigned to test vessels?	Yes
Biological observations made every 24 hr?	Yes
Water Parameter Measurements 1. Temperature: Measured constantly or, if water baths are used, every 6 hr, may not vary > 1°C 2. DO and pH: Measured at beginning of test and every 48 hr in the high, medium, and low doses and in the control	1. Temperature was measured in all replicates and reported every 24 hr for vessels with surviving fish (Appendix 1). It did not vary more than 1°C. 2. DO and pH were measured in all replicates and reported every 24 hr for vessels with surviving fish (Appendix 1).

Guideline Criteria	Reported Information
Chemical Analysis Needed if solutions were aerated, if chemical was volatile, insoluble, or known to absorb, if precipitate formed, if containers were not steel or glass, or if flow-through system was used	Yes. Water samples were taken in duplicate from the control and each exposure level (samples from replicate vessels were pooled) at 0, 48, and 72 hr (fresh media) and at 24, 72, and 96 hr (expired media). No aeration was provided.

12. REPORTED RESULTS

A. General Results

Guideline Criteria	Reported Information
Quality assurance and GLP compliance statements were included in the report?	Yes
Recovery of Chemical	Not reported
Control Mortality Not more than 10% control organisms may die or show abnormal behavior.	0%
Raw data included?	Yes
Signs of toxicity (if any) were described?	Yes

Mortality[4]

Concentration (ppm)		Number of Fish	Cumulative Number Dead			
			Hour of Study			
Nominal	Mean Measured		24	48	72	96
Control	--	20	0	0	0	0
Solvent Control	--	N/A	N/A	N/A	N/A	N/A
1.0	--	20	0	0	0	0
1.8	--	20	0	6	11	13
3.2	--	20	20	20	20	20

[4] All concentrations were reported in mg/L, which the technical reviewer converted to the ppm equivalents. While measured concentrations were reported, the authors reported all results in terms of nominal concentrations.

Concentration (ppm)		Number of Fish	Cumulative Number Dead			
			Hour of Study			
Nominal	Mean Measured		24	48	72	96
5.6	--	20	20	20	20	20
10.0	--	20	20	20	20	20

Other significant results: Fish exhibited non-lethal effects from 3 to 6 hrs after test initiation at the 3.2 ppm and 5.6 ppm levels (all fish were dead at 10 ppm after only 3 hrs), including lying on the bottom of the vessels, increased pigmentation, and loss of equilibrium.

B. Statistical Results[5]

Method: LC_{50}, logistic model (Berkson, 1944[6]); 95% C.I., likelihood ratio method (Williams, 1986[6]); NOEL, the highest concentration tested at which no significant mortality or sublethal adverse effects were observed (an incidence rate of more than one affected fish out of ten, in any replicate, was considered significant).

96-hr LC_{50}: 1.7 ppm (nominal) 95% C.I.: 1.5 - 1.8 ppm (nominal)
Probit Slope: N/A NOEC: 1.0 ppm

Other results (nominal):
3-hr LC_{50} (95% C.I.): 5.1 (4.4 - 5.4) ppm
6-hr LC_{50} (95% C.I.): 3.7 (3.4 - 4.3) ppm
24-hr LC_{50} (95% C.I.): 2.4 (2.1 - 2.8) ppm
48-hr LC_{50} (95% C.I.): 1.9 (1.8- 2.2) ppm
72-hr LC_{50} (95% C.I.): 1.8 (1.6 - 1.9) ppm

13. VERIFICATION OF STATISTICAL RESULTS

Parameter	Result (measured)
Binomial Test LC_{50} (C.I.)	Not used
Moving Average Angle LC_{50} (95% C.I.)	Not used
Probit LC_{50} (95% C.I.)	Not appropriate
Probit Slope	Not appropriate

[5] All concentrations were reported in mg/L, which the technical reviewer converted to the ppm equivalents.

[6] Berkson, J. 1944. Application of the logistic function to bio-assay. *J. Amer. Statist. Assoc.* 39: 357-365.

Williams, D.A. 1986. Interval estimation of the median lethal dose. *Biometrics* 42:641-645.

Parameter	Result (measured)
NOEC (Dunnett's test)	0.58 ppm
96-hr Spearman-Karber LC_{50} (95% C.I.)	0.87 (0.79 - 0.96) ppm

Comments: To verify the statistical results, we used the mean measured concentrations as described in Section 14.

14. REVIEWER'S COMMENTS

All results reported by the authors were in terms of nominal concentrations. Chemical analysis of fresh media (samples taken after media renewal at 0, 48, and 72 hr) and expired media (samples taken from the same "used" media at 24, 72, and 96 hr) revealed that the measured concentrations ranged from 53% to 89% of nominal for the fresh media and 30% to 67% for the expired media. The authors noted that while near nominal concentrations were achieved for the fresh media, difficulties in maintaining homogeneity and stability of NOVARON accounted for the losses in the expired media.

To verify the statistical results, we used the mean measured concentrations, as shown in the table below:

Concentrations of Zirconium Phosphate as Measured by a Turbidimetric Method in the Lowest Three Exposure Groups

Hour Sample Taken	Fresh or Expired Solution[a]	Nominal Concentration (ppm)		
		1.0	1.8	3.2
		Measured Concentration (ppm)		
0	fresh	0.89	1.36	1.71
24	expired	0.67	0.89	1.18
48	fresh	0.58	1.14	--
72	expired	0.39	0.54	--
72	fresh	0.54	1.02	--
96	expired	0.43	0.77	--
Average		0.58	0.95	1.44

[a] Expired solutions had been in test vessels for 24 hr.

As described in Section 14 of the review for MRID No. 445829-18, we do not believe that the turbidimetric method of analyzing the test water for zirconium phosphate was adequate, however.

Also as described in Section 14 of the review for MRID No. 445829-18, we do not understand how zirconium phosphate could be used to assess the aquatic toxicity of AlphaSan™ RC 5000, which contains 3.8% silver by weight. Silver is extremely toxic to aquatic organisms.

DATA EVALUATION REPORT

ZIRCONIUM PHOSPHATE
(NOVARON AG300)

Study Type: Acute LC_{50} Test with a Freshwater Invertebrate (*Daphnia magna*)

Prepared for

Antimicrobial Division
Office of Pesticide Programs
U.S. Environmental Protection Agency
1921 Jefferson Davis Highway
Arlington, VA 22202

Prepared by

ICF Consulting Group
9300 Lee Highway
Fairfax, VA 22031

Principal Reviewer	Matthew C. Preisser *Matthew Preisser, M.E.M.*	Date 3/31/99
Independent Reviewer	Margaret McVey *Margaret McVey, Ph.D.*	Date 3/31/99
Project Manager (QA/QC Manager)	Ellen Mantus *Ellen Mantus, Ph.D.*	Date 4/5/99

Contract Number: 68-W6-0022
Work Assignment No.: 3-17
EPA Project Manager: Peter Thompson

Disclaimer

This review may have been changed following contractor's submission to the Antimicrobial Division of the Office of Pesticide Programs.

DP Barcode: D253522

MRID No.: 445829-20

DATA EVALUATION RECORD
§ 72-2 -- ACUTE LC_{50} TEST WITH A FRESHWATER INVERTEBRATE

1. CHEMICAL: Zirconium phosphate[1]

PC Code No.:

2. TEST MATERIAL: NOVARON AG300
(Batch No. 7130519; white powder)

Purity: ≥ 99%

3. CITATION

Authors[2]:	G. Bell, S. Groom, and B. Smith
Title:	Application for Pesticide Registration: *AlphaSan™ RC 5000*, EPA File Symbol 11631- , Volume 22: Acute Toxicity to *Daphnia Magna*
Study Completion Date:	July 13, 1995
Laboratory:	Huntingdon Research Centre, Ltd., P.O. Box 2, Huntingdon, Cambridgeshire, PE18 6ES, England
Sponsor:	Milliken Chemical, A Division of Milliken & Company, P.O. Box 1927, Spartanburg, SC 29304-1927
Laboratory Report ID:	TSI 64(b)/950718
MRID No.:	445829-20
DP Barcode:	D253522

4. REVIEWED BY: W. Jakob, Antimicrobials Division (7510C)

Signature: Wanda J Jakob Date: 08/09/99

5. APPROVED BY: P. Thompson, Ph.D., Antimicrobials Division (7510C)

Signature: Wanda J Jakob Date: 08/09/99

6. STUDY PARAMETERS

Age of Test Organisms:	1st instar (< 24 hr old)
Definitive Test Duration:	48 hr (02/15/95 to 02/17/95)
Study Method:	Static
Type of Concentrations:	Nominal

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[1] The acute toxicity test report is being submitted in support of the reregistration of AlphaSan™ RC 5000, which contains silver sodium hydrogen zirconium phosphate (3.8% silver by weight). The toxicity test report under review indicates that the test substance was NOVARON AG300, which is ≥ 99% "partially ion-exchange" zirconium phosphate. There is no evidence that the test substance contained any silver, which is the true active ingredient.

[2] The authors were the personnel responsible for conducting the experiments and were affiliated with the performing laboratory, Huntingdon Research Centre, Ltd. The MRID report was prepared by Keller and Heckman LLP, 1001 G Street, N.W., Suite 500 West, Washington, D.C., 20001.

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7. CONCLUSIONS

Results Synopsis[3]:
EC_{50} (immobilization): 0.13 ppm 95% C.I.: 0.12 - 0.16 ppm
NOEC: Not reported Probit Slope: N/A

8. ADEQUACY OF THE STUDY

A. Classification: Invalid

B. Rationale: According to information provided on page 10 of the report, the test substance did not contain silver, which is one of the constituents of the substance being submitted for reregistration. Furthermore, measured concentrations ranged from 87% to 127% of nominal at 0 hr but were only 0% to 15% of nominal at 48 hr. It is possible that the test substance was slowly hydrating and "dissolving" into the water so that after 48 hr it could no longer be detected by the turbidimetric method employed. Basically, there is no way to determine the exposure concentrations for the active ingredient (silver) or the zirconium phosphate.

C. Repairability: Request from sponsor information on whether the test substance supplied to the test laboratory contained silver and if so, how much silver was in the test substance.

9. GUIDELINE DEVIATIONS

1. The authors did not report all of the required acclimation information, including the length of the acclimation period, pretest mortality, health of the test organisms, or the presence of ephippia.

2. The hardness of the dilution water was not reported.

3. The reported pH range (7.6 to 8.3) was above the recommended pH range given in these § 72-2 guidelines (7.2 to 7.6). The EPA 1996 harmonized guidelines (OPPTS 850.1010) do not establish a recommended pH range.

4. A range-finding test was not conducted or data was not reported.

5. Percent recovery of the chemical was not reported.

6. A NOEC was not reported despite adequate data to determine the NOEC for immobilization.

10. SUBMISSION PURPOSE: Reregistration

[3] Study results reported in mg/L and converted to ppm by the technical reviewer.

11. MATERIALS AND METHODS

A. Test Organisms

Guideline Criteria	Reported Information
Species Preferred species is *Daphnia magna*	*Daphnia magna*
All organisms are approximately the same size and weight?	Not reported
Life Stage Daphnids: 1st instar (< 24 hr) Amphipods, stoneflies, and mayflies: 2nd instar Midges: 2nd & 3th instar	1st instar (< 24 hr)
Supplier	Huntingdon Research Centre, Ltd., laboratory culture originating from a strain supplied by the Institute National de Recherche Chimique Appliquée (IRChA), France
All organisms from the same source?	Yes

B. Source/Acclimation

Guideline Criteria	Reported Information
Acclimation Period Minimum 7 days	Brood stocks cultured under test conditions for an unreported length of time; gravid adults were isolated 24 hr prior to test initiation
Wild caught organisms were quarantined for 7 days?	N/A
Were there signs of disease or injury?	Not reported
If treated for disease, was there no sign of the disease remaining during the 48 hr prior to testing?	N/A
Feeding No feeding during the study	No feeding during the study
Pretest Mortality No more than 3% mortality 48 hr prior to testing	Not reported

C. Test System

Guideline Criteria	Reported Information
Source of dilution water Soft reconstituted water or water from a natural source, *not* dechlorinated tap water	Elendt M7 medium, prepared using analytical grade reagents and reverse osmosis purified water (see Appendix 3 of the report)
Does water support test animals without observable signs of stress?	Not reported
Water Temperature Daphnia: 20 °C Amphipods and mayflies: 17 °C Midges and mayflies: 22 °C Stoneflies: 12 °C	21 °C
pH Prefer 7.2 to 7.6	pH range: 7.6 - 8.3
Dissolved Oxygen Static: ≥ 60% during 1st 48 hr and ≥ 40% during 2nd 48 hr; flow-through: ≥ 60%	DO range: 7.6 to 8.4 mg/L (approximately 85% to 95% saturation at sea level and 21 °C)
Total Hardness Prefer 40 to 48 mg/L as $CaCO_3$	Not reported
Test Aquaria 1. Material: Glass or stainless steel 2. Size: 250 mL (daphnids and midges) or 3.9 L (1 gal) 3. Fill volume: 200 mL (daphnids and midges) or 2-3 L	1. Material: Glass 2. Size: Not reported 3. Fill volume: 250 mL
Type of Dilution System Must provide reproducible supply of toxicant	N/A
Flow Rate Consistent flow rate of 5-10 vol/24 hr; meter systems calibrated before study and checked twice daily during test period	N/A
Biomass Loading Rate Static: ≤ 0.8 g/L at ≤ 17°C, ≤ 0.5 g/L at > 17°C; flow-through: ≤ 1 g/L/day	25 mL test solution/organism
Photoperiod 16 hr light, 8 hr dark	16 hr light, 8 hr dark

Guideline Criteria	Reported Information
Solvents Not to exceed 0.5 ml/L for static tests or 0.1 ml/L for flow-through tests	None

D. Test Design

Guideline Criteria	Reported Information
Range Finding Test If LC_{50} >100 mg/L, then no definitive test is required	Not reported
Nominal Concentrations of Definitive Test Control & 5 treatment levels; a geometric series with each concentration being at least 60% of the next higher one	Negative control and 5 nominal test concentrations (ppm): 0.10; 0.18; 0.32; 0.56; and 1.0; each concentration is 56% of the next higher concentration; concentrations were in a geometric series
Number of Test Organisms Minimum 20/level, may be divided among containers	20/level (10 test organisms per test vessel and 2 test vessels per level)
Test organisms randomly or impartially assigned to test vessels?	Yes
Water Parameter Measurements 1. Temperature: Measured continuously or, if water baths are used, every 6 hr; may not vary > 1 °C 2. DO and pH: Measured at beginning of test and every 48 hr in the high, medium, and low doses and in the control	1. Temperature in each vessel was measured daily (i.e., at 0 hr, 24 hr, and 48 hr) and did not vary by more than 1 °C 2. DO and pH were measured in each vessel at the start and end of the study (i.e., at 0 hr and 48 hr)
Chemical Analysis Needed if solutions were aerated, if chemical was volatile, insoluble, or known to absorb, if precipitate formed, if containers were not steel or glass, or if flow-through system was used	Yes. Water samples were taken from each control and exposure vessel (data were pooled) at 0 hr and 48 hr. Solutions were not aerated.

12. REPORTED RESULTS

Guideline Criteria	Reported Information
Quality assurance and GLP compliance statements were included in the report?	Yes

Guideline Criteria	Reported Information
Control Mortality Static: ≤ 10% Flow-through: ≤ 5%	0%
Percent Recovery of Chemical	Not reported
Raw data included?	Yes

A. Mortality[4]

Concentration (ppm)		Number of Organ-isms	Cumulative Number Dead			
			Hour of Study			
Nominal	Mean Measured		24	48	72	96
Control	--	20	0	0	0	0
Solvent Control	--	N/A	N/A	N/A	N/A	N/A
0.10	0.06	20	0	0	N/A	N/A
0.18	0.085	20	20	20	N/A	N/A
0.32	0.14	20	20	20	N/A	N/A
0.56	0.36	20	20	20	N/A	N/A
1.0	0.64	20	20	20	N/A	N/A

B. Statistical Results[4]

Method: EC_{50}, logistic model (Berkson, 1944[5]); 95% C.I., liklihood ratio method (Williams, 1986[5])

48-hr EC_{50} (immobilization): 0.13 ppm 95% C.I.: 0.12 - 0.16 ppm
Probit Slope: N/A NOEC: Not reported

[4] All concentrations were reported in mg/L, which the technical reviewer converted to the ppm equivalents. Mean measured concentrations were calculated by the technical reviewer based on the reported adjusted measured concentrations at 0 hr and 48 hr (see Appendix 2, Table C1 of the report).

[5] Berkson, J. 1944. Application of the logistic function to bio-assay. *J. Amer. Statist. Assoc.* 39: 357-365.

Williams, D.A. 1986. Interval estimation of the median lethal dose. *Biometrics* 42:641-645.

Other results:

24-hr EC_{50} (immobilization): 0.13 ppm 95% C.I.: 0.12 - 0.16 ppm

13. VERIFICATION OF STATISTICAL RESULTS

Parameter	Result
Binomial Test EC_{50} (95 % C.I.)	Not used
Moving Average Angle LC_{50} (95% C.I.)	Not used
Probit LC_{50} (95% C.I.)	Not appropriate
Probit Slope	Not appropriate
Graphic Method LC_{50}	0.071 ppm
NOEC	0.06 ppm

14. REVIEWER'S COMMENTS

All results reported by the authors are expressed in terms of nominal concentrations. Chemical concentrations measured using a turbidimetric method ranged from 87% to 127% of nominal at 0 hr to 0% to 15% of nominal at 48 hr. The authors noted that while near nominal concentrations were achieved for the fresh media, difficulties in maintaining test material stability accounted for the decrease in measured concentrations recorded at 48 hr. They did note that the test vessels were loosely covered with aluminum foil to minimize evaporation loss. The zirconium phosphate might simply have become hydrated and "dissolved" in the water over time so that a turbidimetric method would no longer detect it. If that is true, then the nominal concentrations would be more appropriate to estimate the LC_{50} and NOEC values than the "measured" concentrations. If that is true, a new difficulty would be that the test substance was not dissolved properly at the beginning of the test (and was never dissolved properly in the tests reported under MRID Nos. 445829-18 and -19).

As noted in our review of MRID No. 445829-18, we do not understand how zirconium phosphate alone could be used to assess the acute aquatic toxicity of AlphaSan™ RC 5000, which contains 3.8% silver by weight. Silver is extremely toxic to aquatic organisms. The proposed freshwater acute ambient water quality criterion for silver is 0.92 μg/L (ppt).

DATA EVALUATION REPORT

Experimental Additive 9823-37

Study Type: Avian Single-Dose LD_{50} Test (*Colinus virginianus*)

Prepared for

Antimicrobial Division
Office of Pesticide Programs
U.S. Environmental Protection Agency
1921 Jefferson Davis Highway
Arlington, VA 22202

Prepared by

ICF Consulting Group
9300 Lee Highway
Fairfax, VA 22031

Principal Reviewer	Matthew C. Preisser *Matthew Preisser, M.E.M.*	Date 3/31/99
Independent Reviewer	Margaret E. McVey *Margaret McVey, Ph.D.*	Date 3/31/99
Project Manager (QA/QC Manager)	Ellen Mantus *Ellen Mantus, Ph.D.*	Date 4/5/99

Contract Number: 68-W6-0022
Work Assignment No.: 3-17
EPA Project Manager: Peter Thompson

Disclaimer

This review may have been changed following contractor's submission to the Antimicrobial Division of the Office of Pesticide Programs.

DP Barcode: D253522 MRID No.: 445829-21

DATA EVALUATION RECORD
§ 71-1(A) - AVIAN SINGLE-DOSE LD_{50} TEST

1. CHEMICAL: Not reported[1] PC Code No.:

2. TEST MATERIAL: Experimental Additive 9823-37 (Lot No. 7170706; white powder) Purity: 100%

3. CITATION

Author[2]:	B.R. Helsten (study director)
Title:	Avian Acute Oral Toxicity Test in Bobwhite Quail with Experimental Additive 9823-37
Study Completion Date:	March 17, 1998
Laboratory:	Bio-Life® Associates, Ltd. (BLAL), N6230 County Road G, Neillsville, WI 54456-8773
Sponsor:	Milliken Chemical, A Division of Milliken & Company, P.O. Box 1927, Spartanburg, SC 29304-1927
Laboratory Report ID:	BLAL No. 163-001-03
DP Barcode:	D253522
MRID No.:	445829-21

4. REVIEWED BY: W. Jakob, Antimicrobials Division (7510C)

Signature: Wanda J Jakob Date: 08/09/99

5. APPROVED BY: P. Thompson, Ph.D., Antimicrobials Division (7510C)

Signature: Wanda J Jakob Date: 08/09/99

6. STUDY PARAMETERS

Scientific Name of Test Organism:	*Colinus virginianus*
Test Organisms Age/Size:	Age: 21 weeks Size: 221.0 to 254.1 g
Definitive Study Duration:	14 days (02/17/98 to 03/03/98)

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[1] The acute toxicity test report is being submitted in support of reregistration of AlphaSan™ RC 5000, which contains silver sodium hydrogen zirconium phosphate. The study report provides no evidence that the test material contained the substance proposed for reregistration. The silver content of 9.9% by weight in the test material suggests that the test material might have been AlphaSan™ RC 2000, but the report lacks the appropriate documentation from the Milliken Chemical Company or Bio-Life® Associates.

[2] The authors were the personnel responsible for conducting the experiments and were affiliated with the performing laboratory, Bio-Life® Associates, Ltd. The MRID report was prepared by Keller and Heckman LLP, 1001 G Street, N.W., Suite 500 West, Washington, D.C., 20001.

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7. CONCLUSIONS

Results Synopsis:
LD_{50}: > 2000 mg/kg
NOEC (mortality): 2000 mg/kg
95% C.I.: N/A
Probit Slope: N/A

8. ADEQUACY OF THE STUDY

A. Classification: Supplemental

B. Rationale: The authors did not document the identity of the test substance or whether it is equivalent to AlphaSan™ RC 5000 (or AlphaSan™ RC 2000).

C. Repairability: Request information on the chemical composition of the test substance from Milliken Chemical Associates, Ltd.

9. GUIDELINE DEVIATIONS

1. The chemical name was not provided in the report or accompanying appendices.

2. Only one test dose was used, but EPA's 1996 draft harmonized guidelines (OPPTS 850.2100) state that "for test substances expected to have relatively low toxicity, a limit test may be conducted at 2000 mg/kg body weight" and, if the definitive test guidelines are followed, "no further testing is required at lower dosage levels."

3. Cage ceilings and walls were open wire mesh, which could allow cross-contamination of feed. However, given the lack of toxicity at 2,000 mg/kg, this is not an important deviation.

10. SUBMISSION PURPOSE: Reregistration

11. MATERIALS AND METHODS

A. Test Organisms

Guideline Criteria	Reported Information
Species A wild waterfowl species, preferably the mallard (*Anas platyrhynchos*), or an upland game bird species, preferably the bobwhite (*Colinus virginianus*)	Bobwhite quail (*Colinus virginianus*)
Age at beginning of test At least 16 weeks old	21 weeks old
Supplier	Circle 7 Quail Farm, 1309 Reynolds Street, Springhill, LA 71075

Guideline Criteria	Reported Information
Acclimation period At least 15 days	19 days

B. Test System

Guideline Criteria	Reported Information
Pen facilities adequate?	Yes. Five birds per pen, each constructed entirely of steel wire mesh, measuring 51 x 51 x 25 cm in size, and maintained over steel pans to collect feces. The OPPTS 850.2100 guidlines recommend that solid sheeting be used for common walls and ceilings.
Photoperiod 10 hr light, 14 hr dark is recommended	8 hr light, 16 hr dark (using natural daylight spectrum lights), which is recommended by the OPPTS 850.2100 guidlines
Diet was nutritious and appropriate for species?	Yes. During acclimation, the birds were fed food treated with Bacitracin Methylene Disalicylate and water treated with Lincomycin-Spectinomycin; untreated food and water was offered during the definitive study; water quality analysis revealed no known contaminants.
Feed withheld at least 15 hr prior to dosing?	Birds were fasted 17.75 hr prior to dosing (water was still available)

Other reported information: room temperature (recorded daily) ranged from 21 to 24 °C and relative humidity ranged from 48% to 56% during the study.

C. Test Design

Guideline Criteria	Reported Information
Range finding test?	Yes (10-day range finding test conducted from 02/11/98 to 02/20/98 at dose levels of 500, 1000, and 2000 mg/kg; a single mortality was recorded at 2000 mg/kg)

Guideline Criteria	Reported Information
Definitive Test Nominal concentrations: At least five, in a geometric scale, unless LD_{50} > 2000 mg ai / kg Controls: Water control or vehicle control (if vehicle is used)	Nominal concentrations: One nominal concentration (single dose): 2000 mg/kg Control birds received empty gelatin capsules
Number of birds per group 10 (strongly recommended)	10 per group (5 males and 5 females)
Vehicle Distilled water, corn oil, propylene glycol, 1% carboxymethylcellulose, or gum arabic	Each bird received its single dose in two gelatin capsules
Amount of vehicle per body weight Constant volume/weight % of body weight, not to exceed 1% (1ml/100g)	N/A
Observations period At least 14 days	14 days

12. REPORTED RESULTS

Guideline Criteria	Reported Information
Quality assurance and GLP compliance statements were included in the report?	Yes
Individual body weights measured at beginning of test, on day 14 and at end of test if extended beyond 14 days?	Individual body weights measured just prior to dosing and on Days 3, 7, and 14
Mean feed consumption measured at beginning of test, on day 14, and at end of test if extended beyond 14 days?	Group feed consumption measured on Days 3, 7, and 14
Control Mortality Not more than 10%	0%
Raw data included?	Yes (mortality, body weight, food consumption)
Signs of toxicity (if any) were described?	Yes

Mortality

Dosage (mg/kg)	No. of Birds	Cumulative Number of Dead							
		Day of Study							
		1	2	3	4	5	6-8	9-11	12-14
Control	10	0	0	0	0	0	0	0	0
2000	10	0	0	0	0	0	0	0	0

Other significant results: There were no significant differences in body weight or food consumption between the test group and the control group.

Sublethal effects included diarrhea in the 2000 mg/kg test group approximately 7.5 hr after dosing on Day 1 and later on Day 3.

After the completion of the study, four randomly selected birds (two males and two females) from each of the control and 2000 mg/kg test groups were subjected to complete necropsy examinations. One female control bird had a pale, mottled liver. A gallbladder enlarged 1.25 times was noted in one female 2000 mg/kg test bird and gaseous intestines were noted in one male 2000 mg/kg test bird.

Reported Statistical Results

Statistical Method: Visual inspection of the data

LD_{50}: > 2000 mg/kg
NOEL (mortality): 2000 mg/kg

95% C.I.: N/A
Probit Slope: N/A

13. VERIFICATION OF STATISTICAL RESULTS

Statistical Method: Visual inspection of the data

LD_{50}: > 2000 mg/kg
NOEL (mortality): ≥ 2000 mg/kg

95% C.I.: N/A
Probit Slope: N/A

Adjusted for 100% active ingredient (optional if > 80% ai): Not necessary (purity = 100%)

15. REVIEWER'S COMMENTS

The acute toxicity test report is being submitted in support of reregistration of AlphaSan™ RC 5000, which contains silver sodium hydrogen zirconium phosphate. The study report provides no evidence that the test material contained the substance proposed for reregistration. The silver content of 9.9% by weight in the test material suggests that the test material might have been AlphaSan™ RC 2000, but the report lacks the appropriate documentation from the Milliken Chemical Company or Bio-Life® Associates. ← ?

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