

US EPA ARCHIVE DOCUMENT

MRID No. 443322-29

DATA EVALUATION RECORD
§ 72-2 - ACUTE EC₅₀ TEST WITH A FRESHWATER INVERTEBRATE

1. **CHEMICAL:** S-Dimethenamid PC Code No.: 120051
2. **TEST MATERIAL:** SAN 1289H Purity:
 Technical 91.1% (S-dimethenamid)
96.3% (tot. dimethenamid)
3. **CITATION:**
Author: William C. Graves and James P. Swigert
Title: SAN 1289H Technical: A 48-Hour Flow-Through Acute Toxicity Test with the Cladoceran (*Daphnia magna*)
Study Completion Date: June 4, 1996
Laboratory: Wildlife International Ltd., Easton, MD
Sponsor: Sandoz Agro, Inc., Des Plaines, IL
Laboratory Report ID: 131A-164
MRID No.: 443322-29
DP Barcode: D238350, D238356

4. **REVIEWED BY:** Karl Bullock, M.S., Associate Scientist,
 Golder Associates, Inc.

Signature: *Karl Bullock* **Date:** 10/24/97

APPROVED BY: Pim Kosalwat, Ph.D., Senior Scientist,
 Golder Associates, Inc.

Signature: *P. Kosalwat* **Date:** 10/24/97

5. **APPROVED BY:**

Signature: *Joan Edwards* **Date:** 12/3/97
Jim C. Bulby 11/9/98

6. **STUDY PARAMETERS:**

Age of Test Organism: <24 hours
Definitive Test Duration: 48 hours
Study Method: Flow-through
Type of Concentrations: Mean measured as total dimethenamid

7. **CONCLUSIONS:** This study is scientifically sound and fulfills the guideline requirements. The 48-hour EC₅₀ value of 12 ppm classifies SAN 1289H technical as slightly toxic to *Daphnia magna*. The NOEC was determined to be 3.4 ppm.



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Results Synopsis

EC₅₀: 12 ppm
 NOEC: 3.4 ppm

95% C.I.: 10-13 ppm
 Probit Slope: 15

8. ADEQUACY OF THE STUDY:

A. **Classification:** Core.

B. **Rationale:** N/A.

C. **Repairability:** N/A.

9. **GUIDELINE DEVIATIONS:** The pH of the dilution water was greater than recommended. This deviation is not believed to detract from the validity of the study.

10. SUBMISSION PURPOSE:**11. MATERIALS AND METHODS:****A. Test Organisms**

Guideline Criteria	Reported Information
<u>Species</u> Preferred species is <i>Daphnia magna</i>	<i>Daphnia magna</i>
All organisms are approximately the same size and weight?	Not reported
<u>Life Stage</u> Daphnids: 1 st instar (<24 h). Amphipods, stoneflies, and mayflies: 2 nd instar. Midges: 2 nd & 3 rd instar.	1 st instar (<24 h)
<u>Supplier</u>	In-house cultures
All organisms from the same source?	Yes

B. Source/Acclimation

Guideline Criteria	Reported Information
<u>Acclimation Period</u> Minimum 7 days	Cultures were maintained under conditions similar to testing.
Wild caught organisms were quarantined for 7 days?	N/A
Were there signs of disease or injury?	No signs of sickness or injury were observed
If treated for disease, was there no sign of the disease remaining during the 48 hours prior to testing?	N/A
<u>Feeding</u> No feeding during the study.	No feeding during the study
<u>Pretest Mortality</u> No more than 3% mortality 48 hours prior to testing.	Not reported

C. Test System

Guideline Criteria	Reported Information
<u>Source of dilution water</u> Soft reconstituted water or water from a natural source, not dechlorinated tap water.	Well water, filtered and aerated before use.
Does water support test animals without observable signs of stress?	Yes
<u>Water Temperature</u> Daphnia: 20°C Amphipods and mayflies: 17°C Midges and mayflies: 22°C Stoneflies: 12°C	19.8-20.0°C
<u>pH</u> Prefer 7.2 to 7.6.	8.1-8.3
<u>Dissolved Oxygen</u> Static: ≥ 60% during 1 st 48 h and ≥ 40% during 2 nd 48 h, flow-through: ≥ 60%.	≥60% of saturation during the test

Guideline Criteria	Reported Information
<p><u>Total Hardness</u> Prefer 40 to 200 mg/L as CaCO₃.</p>	136 mg/L as CaCO ₃
<p><u>Test Aquaria</u> 1. <u>Material:</u> Glass or stainless steel. 2. <u>Size:</u> 250 mL (daphnids and mides) or 3.9 L (1 gal). 3. <u>Fill volume:</u> 200 mL (daphnids and mides) or 2-3 L.</p>	Each test chamber was a 300-mL glass beaker, with Nytex [®] screen covering both ends. The beakers were suspended in an 8-L stainless steel chamber filled with 6.5 L of test solution.
<p><u>Type of Dilution System</u> Must provide reproducible supply of toxicant.</p>	Continuous-flow diluter
<p><u>Flow Rate</u> Consistent flow rate of 5-10 vol/24 hours, meter systems calibrated before study and checked twice daily during test period.</p>	14 vol/24 hours; meter systems calibrated before the test and checked twice daily during the test.
<p><u>Biomass Loading Rate</u> Static: ≤ 0.8 g/L at ≤ 17°C, ≤ 0.5 g/L at > 17°C; flow-through: ≤ 1 g/L/day.</p>	Not reported
<p><u>Photoperiod</u> 16 hours light, 8 hours dark.</p>	16 hours light, 8 hours dark
<p><u>Solvents</u> Not to exceed 0.5 mL/L for static tests or 0.1 mL/L for flow-through tests.</p>	Solvent: dimethylformamide Maximum conc.: 0.10 ml/L

D. Test Design

Guideline Criteria	Reported Information
<p><u>Range Finding Test</u> If EC₅₀ >100 mg/L, then no definitive test is required.</p>	No
<p><u>Nominal Concentrations of Definitive Test</u> Control & 5 treatment levels; a geometric series with each concentration being at least 60% of the next higher one.</p>	Negative control, solvent control, 3.2, 5.4, 9.0, 15, and 25 mg/L, not corrected for purity
<p><u>Number of Test Organisms</u> Minimum 20/level, may be divided among containers.</p>	20 per level, 10 per replicate
<p>Test organisms randomly or impartially assigned to test vessels?</p>	Yes
<p><u>Water Parameter Measurements</u></p> <ol style="list-style-type: none"> <u>Temperature</u> Measured continuously or, if water baths are used, every 6 h, may not vary > 1°C. <u>DO and pH</u> Measured at beginning of test and ever 48 h in the high, medium, and low doses and in the control. 	<p>Temperature measured in each chamber at test initiation and termination, and also measured continuously in one negative control replicate</p> <p>DO and pH measured at test initiation and every 24 hours in alternate replicate test chambers</p>
<p><u>Chemical Analysis</u> 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</p>	Solutions collected from each replicate test chamber at test initiation and termination and analyzed by GC-NPD

13. VERIFICATION OF STATISTICAL RESULTS:

Parameter	Result
Binomial Test EC ₅₀ (95% C.I.)	11 ppm (9-14 ppm)
Moving Average Angle EC ₅₀ (95% C.I.)	12 ppm (10-14 ppm)
Probit EC ₅₀ (95% C.I.)	12 ppm (10-13 ppm)
Probit Slope	15
NOEC	3.4 ppm

- 14. REVIEWER'S COMMENTS:** This study is scientifically sound, fulfills the guideline requirements, and can be classified as Core. The 48-hour EC₅₀ for *Daphnia magna* exposed to SAN 1289H was determined to be 12 ppm, which classifies this compound as slightly toxic to the daphnid. The NOEC was determined to be 3.4 ppm.

KARL BULLOCK S-DIMETHENAMID DAPHNIA 10-21-97

CONC.	NUMBER EXPOSED	NUMBER DEAD	PERCENT DEAD	BINOMIAL PROB. (PERCENT)
26	20	20	100	9.536742E-05
14	20	18	90	2.012253E-02
9	20	1	5	2.002716E-03
5.2	20	0	0	9.536742E-05
3.4	20	0	0	9.536742E-05

THE BINOMIAL TEST SHOWS THAT 9 AND 14 CAN BE USED AS STATISTICALLY SOUND CONSERVATIVE 95 PERCENT CONFIDENCE LIMITS, BECAUSE THE ACTUAL CONFIDENCE LEVEL ASSOCIATED WITH THESE LIMITS IS GREATER THAN 95 PERCENT.

AN APPROXIMATE LC50 FOR THIS SET OF DATA IS 11.4451

RESULTS CALCULATED USING THE MOVING AVERAGE METHOD
SPAN G LC50 95 PERCENT CONFIDENCE LIMITS
3 5.135012E-02 11.74197 10.22869 13.52779

RESULTS CALCULATED USING THE PROBIT METHOD
ITERATIONS G H GOODNESS OF FIT PROBABILITY
7 .1656869 1 1

SLOPE = 15.25076
95 PERCENT CONFIDENCE LIMITS = 9.042988 AND 21.45853

LC50 = 11.53709
95 PERCENT CONFIDENCE LIMITS = 10.43469 AND 12.66142

LC10 = 9.524047
95 PERCENT CONFIDENCE LIMITS = 7.934804 AND 10.51481

12. REPORTED RESULTS:**A. General Results**

Guideline Criteria	Reported Information
Quality assurance and GLP compliance statements were included in the report?	Yes
Control Mortality Static: ≤10% Flow-through: ≤5%	0% mortality in the control groups
Percent Recovery of Chemical	93-106% of nominal values
Raw data included?	Yes

Immobility

Mean Measured Concentration (ppm) *	Number of Daphnids	Cumulative Number Immobile/Dead	
		24-hr	48-hr
Control	20	0	0
Solvent Control	20	0	0
3.4	20	0	0
5.2	20	0	0
9.0	20	0	1
14	20	1	18
26	20	1	20

* as total dimethenamid

Other Significant Results: Signs of toxicity included lethargy, floating, and immobility. Some or all of these signs were observed at the four highest concentrations.

B. Statistical Results: Results are based on mean measured concentrations as total dimethenamid.
Method: Probit

48-hr EC₅₀: 12 ppm
Probit Slope: 15

95% C.I.: 10-13 ppm
NOEC: 3.4 ppm