

US EPA ARCHIVE DOCUMENT



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

JAN 4 2000

OFFICE OF
PREVENTION, PESTICIDES AND
TOXIC SUBSTANCES

MEMORANDUM

SUBJECT: Alkyl (C10-16) Dimethylamine Oxide: Aquatic Toxicity Studies (D260071)

TO: Velma Noble, PM-31
Regulatory Management Branch I
Antimicrobials Division (7510C)

FROM: Kathryn V. Montague, M.S., Biologist
Risk Assessment and Science Support Branch
Antimicrobials Division (7510C)

THRU: Allen W. Vaughan, Team Leader, Team 3
Risk Assessment and Science Support Branch
Antimicrobials Division (7510C)

Norman Cook, Branch Chief
Risk Assessment and Science Support Branch
Antimicrobials Division (7510C)

Procter and Gamble Company has submitted aquatic toxicity data in support of the registration of Alkyl (C10-16) dimethylamine oxide for use in dishwashing detergent. Summaries of the submitted studies follow:

Acute Toxicity of Procter and Gamble Experimental Chemicals Nos. 001 thru 006, and 011 thru 023 to Bluegill Sunfish (*Lepomis macrochirus* Rafinesque) (Guideline 850.1075/old 72-1) (MRID #44434911): The LC₅₀ for amine oxide is 6.4 ppm ai (95 % c.i. of 4.8 - 8.6 ppm ai) (moderately toxic). The NOAEC was not reported. Fish exhibited lethargy, hyperventilation, surfacing, loss of equilibrium, and laid on the bottom of the test vessels prior to death. The study does not fulfill guideline requirements and is classified as supplemental. No further testing is required, however, as discussed in the memorandum from AD dated 6/25/98.

Acute Toxicity of 16 Procter and Gamble Experimental Compounds to Bluegill Sunfish (*Lepomis macrochirus*) (Guideline 850.1075/old 72-1) (MRID #44434912): The LC₅₀ for amine oxide is 3.13 ppm ai (95 % c.i. 2.02 - 4.84 ppm ai) (moderately toxic). The NOAEC was 1.2 ppm ai. Fish became dark and lethargic and lost equilibrium prior to death. The study does not fulfill guideline requirements and is classified as supplemental. No further testing is required, however, as discussed in the memorandum from AD dated 6/25/98.

Maki, A.W. 1979. Correlations Between *Daphnia magna* and Fathead Minnow (*Pimephales promelas*) Chronic Toxicity Values for Several Classes of Test Substances. J. Fish Res. Board Can. 36:411-421. Submitted toward fulfillment of Guideline 850.101/old 72-2. (MRID #44434913). This study provided 96-h acute LC₅₀ information, as well as 21-d LC₅₀ and reproduction EC₅₀ values. The 96-h LC₅₀ was 1.01 ppm (95% c.i. of 0.85 - 1.21 ppm) (moderately toxic). The 21-d LC₅₀ was 0.96 ppm (95% c.i. of 0.90 - 1.03 ppm), and the reproductive EC₅₀ s were 0.88 (0.77 - 1.04), 1.01 (0.95 - 1.07), and 1.04 (0.98 - 1.11) ppm for total young production, average brood size, and percentage of days reproduction occurred, respectively. The overall NOAEC was 0.70 ppm. This study was not conducted according to Guideline criteria, and it does not fulfill Guideline requirements. It is classified as supplemental. No further testing is required, however, as discussed in the memorandum from AD dated 6/25/98.

No additional ecological effects testing is required for the currently proposed uses of alkyl (C10-16) dimethylamine oxide as an ingredient in dishwashing detergent.

If you have any questions on the above, please contact Kathryn Montague (703-305-1243).

DATA EVALUATION RECORD
§ 72-1(A) -- ACUTE LC₅₀ TEST WITH A WARMWATER FISH

1. **CHEMICAL:** Alkyl(C10-16) dimethylamine oxide PC Code: 000439
2. **TEST MATERIAL:** P&G Exper. Chem. 011 Purity: NR

3. **CITATION**

Authors: Hutchinson, C., and K. Macek
Title: Acute Toxicity of Procter & Gamble Experimental Chemicals
Nos. 001 thru 006, and 011 thru 023 to Bluegill Sunfish
(Lepomis macrochirus Rafinesque)
Study Completion Date: April 1970
Laboratory: Bionomics, Inc.
Sponsor: Procter & Gamble
Laboratory Report ID: None reported
MRID No.: 444349-11
DP Barcode: D244070

4. **REVIEWED BY:** Allen W. Vaughan, Entomologist, RASSB/AD

Signature: *Allen W. Vaughan*

Date: 12-27-89

5. **APPROVED BY:** Norman Cook, Branch Chief, RASSB/AD

Signature: *Norman Cook*

Date: 01-04-2000

6. **STUDY PARAMETERS**

Age or Size of Test Organism: Mean wt = 0.7 gm
Mean length = 32 mm
Definitive Test Duration: 96 hours
Study Method: Static
Type of Concentrations: measured

7. **CONCLUSIONS:** This study is scientifically sound, and shows that alkyl dimethylamine oxide is moderately toxic to bluegill sunfish (96-hr LC₅₀ = 6.4 ppm). Because the study deviates significantly from OPP test guidelines, it is considered supplemental.

Results Synopsis

LC₅₀: 6.4 ppm ai

95% C.I.: 4.8-8.6 ppm ai

NOEC: Not reported

Probit Slope: Not reported

8. ADEQUACY OF THE STUDY

A. Classification: Supplemental

B. Rationale: Significant guideline deviations; raw data not provided; test chemical not adequately characterized; data on concentrations of test chemical not provided

C. Repairability: Not repairable

9. GUIDELINE DEVIATIONS

Because of insufficient detail in study report, status of all possible deviations cannot be assessed. One deviation that may be noted in this report is that there were no control (untreated) test organisms.

10. SUBMISSION PURPOSE: Registration

11. MATERIALS AND METHODS

A. Test Organisms

Guideline Criteria	Reported Information
<u>Species</u> Preferred species is the bluegill sunfish <i>(Lepomis macrochirus)</i>	Bluegill sunfish
<u>Mean Weight</u> 0.5-5 g	0.7 g
<u>Mean Standard Length</u> Longest not > 2x shortest	Mean: 32 mm
<u>Supplier</u>	Ozark Fisheries, Stoutland, MO
All fish from same source?	Yes
All fish from the same year class?	not reported

B. Source/Acclimation

Guideline Criteria	Reported Information
Acclimation Period Minimum 14 days	10 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 hours prior to testing?	N/A
Feeding No feeding during the study	Not reported
Pretest Mortality No more than 3% mortality 48 hours prior to testing	<1% mortality prior to testing.

C. Test System

Guideline Criteria	Reported Information
Source of dilution water Soft reconstituted water or water from a natural source, not dechlorinated tap water	Deionized water, source not reported
Does water support test animals without observable signs of stress?	Yes
Water Temperature 17°C or 22°C	18°C
pH Prefer 7.2 to 7.6	7.1
Dissolved Oxygen Static: ≥ 60% during 1 st 48 hrs and ≥ 40% during 2 nd 48 hrs, flow-through: ≥ 60%	4.5-6.0 ppm minimum

Guideline Criteria	Reported Information
<p>Total Hardness Prefer 40 to 48 mg/L as CaCO₃</p>	not reported
<p>Test Aquaria 1. <u>Material</u>: Glass or stainless steel 2. <u>Size</u>: Volume of 19 L (5 gal) or 30 x 60 x 30 cm 3. <u>Fill volume</u>: 15-30 L of solution</p>	Glass, 5 gal., 15 liters of test solution
<p>Type of Dilution System Must provide reproducible supply of toxicant</p>	N/A
<p>Flow Rate Consistent flow rate of 5-10 vol/24 hours, meter systems calibrated before study and checked twice daily during test period</p>	N/A
<p>Biomass Loading Rate Static: ≤ 0.8 g/L at ≤ 17°C, ≤ 0.5 g/L at > 17°C; flow-through: ≤ 1 g/L/day</p>	≤ 1 g/L
<p>Photoperiod 16 hours light, 8 hours dark</p>	Not reported
<p>Solvents Not to exceed 0.5 ml/L for static tests or 0.1 ml/L for flow-through tests</p>	No solvents reported

D. Test Design

Guideline Criteria	Reported Information
<p>Range Finding Test If LC₅₀ > 100 mg/L with 30 fish, then no definitive test is required.</p>	Not reported

Guideline Criteria	Reported Information
<p><u>Nominal Concentrations of Definitive Test</u> Control & 5 treatment levels; dosage should be 60% of the next highest concentration; concentrations should be in a geometric series</p>	Not reported
<p><u>Number of Test Organisms</u> Minimum 10/level, may be divided among containers</p>	10/level
<p><u>Test organisms randomly or impartially assigned to test vessels?</u></p>	Not reported
<p><u>Biological observations made every 24 hours?</u></p>	Yes
<p><u>Water Parameter Measurements</u> 1. <u>Temperature</u> Measured constantly or, if water baths are used, every 6 hrs, may not vary > 1°C 2. <u>DO and pH</u> Measured at beginning of test and every 48 h in the high, medium, and low doses and in the control</p>	Details not reported
<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>	Details not reported

12. REPORTED RESULTS

A. General Results

Guideline Criteria	Reported Information
Quality assurance and GLP compliance statements were included in the report?	No; exempt from GLP since conducted prior to 10/16/89
Recovery of Chemical	Not reported
Control Mortality Not more than 10% control organisms may die or show abnormal behavior.	No negative controls were used
Raw data included?	No
Signs of toxicity (if any) were described?	Yes; fish were lethargic, exhibited hyperventilation and surfacing, and lost equilibrium and laid on the bottom of the test vessels before death.

Other Significant Results:

B. Statistical Results

Method: Details of analysis were not reported.

96-hr LC₅₀: 6.4 ppm ai 95% C.I.: 4.8-8.6 ppm ai

Probit Slope: Not reported NOEC: Not reported

13. VERIFICATION OF STATISTICAL RESULTS

Due to lack of information, no verification was possible.

14. REVIEWER'S COMMENTS: No comments.

DATA EVALUATION RECORD
§ 72-1(A) -- ACUTE LC₅₀ TEST WITH A WARMWATER FISH

1. **CHEMICAL:** Alkyl(C10-16) dimethylamine oxide PC Code:000439

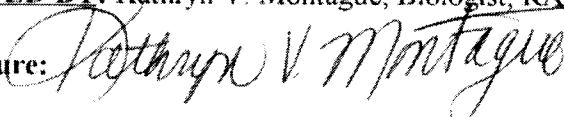
2. **TEST MATERIAL:** P&G Exper. Chem. NPS-72.004 Purity: NR

3. **CITATION**

Authors: Bionomic, Inc.
Title: Acute Toxicity of 16 Procter & Gamble Experimental Compounds to Bluegill Sunfish (Lepomis macrochirus)
Study Completion Date: July 1972
Laboratory: Bionomics, Inc.
Sponsor: Procter & Gamble
Laboratory Report ID: None reported
MRID No.: 444349-12
DP Barcode: D260071

4. **REVIEWED BY:** Kathryn V. Montague, Biologist, RASSB3/AD

Signature:



Date:

12/22/99

5. **APPROVED BY:** Norman Cook, Branch Chief, RASSB/AD

Signature:



Date:

AW 12-27-99
01-04-2000

6. **STUDY PARAMETERS**

Age or Size of Test Organism: Mean wt = 1.2 g, Mean length = 43 mm

Definitive Test Duration: 96 hours

Study Method: Static

Type of Concentrations: Nominal

7. **CONCLUSIONS:** This study is scientifically sound, and shows that alkyl dimethylamine oxide is moderately toxic to bluegill sunfish (96-hr LC₅₀ = 3.13 ppm). Because the study deviates significantly from OPP test guidelines, it is considered supplemental.

Results Synopsis

LC₅₀: 3.13 ppm ai

95% C.I.: 2.02 - 4.84 ppm ai

NOAEC: 1.2 ppm ai

Probit Slope: Not reported

8. ADEQUACY OF THE STUDY

A. Classification: Supplemental

B. Rationale: Significant guideline deviations; raw data not provided; test chemical not adequately characterized; data on concentrations of test chemical not provided

C. Repairability: Not repairable

9. GUIDELINE DEVIATIONS

Because of insufficient detail in study report, status of all possible deviations cannot be assessed. One deviation that may be noted in this report is that there were no control (untreated) test organisms.

10. SUBMISSION PURPOSE: Registration

11. MATERIALS AND METHODS

A. Test Organisms

Guideline Criteria	Reported Information
Species Preferred species is the bluegill sunfish (<i>Lepomis macrochirus</i>)	Bluegill sunfish
Mean Weight 0.5-5 g	1.2 g
Mean Standard Length Longest not > 2x shortest	Mean: 43 mm Range: not reported
Supplier	"a commercial fish hatchery in Nebraska"
All fish from same source?	Yes
All fish from the same year class?	not reported

B. Source/Acclimation

Guideline Criteria	Reported Information
Acclimation Period Minimum 14 days	24 hours
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 hours prior to testing?	N/A
Feeding No feeding during the study	Not reported
Pretest Mortality No more than 3% mortality 48 hours 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	reconstituted deionized water, source not reported
Does water support test animals without observable signs of stress?	not reported
Water Temperature 17°C or 22°C	18°C
pH Prefer 7.2 to 7.6	7.1
Dissolved Oxygen Static: ≥ 60% during 1 st 48 hrs and ≥ 40% during 2 nd 48 hrs, flow-through: ≥ 60%	5.4 - 9.8 mg/L

Guideline Criteria	Reported Information
Total Hardness Prefer 40 to 48 mg/L as CaCO ₃	35 mg/L as CaCO ₃
Test Aquaria 1. <u>Material</u> : Glass or stainless steel 2. <u>Size</u> : Volume of 19 L (5 gal) or 30 x 60 x 30 cm 3. <u>Fill volume</u> : 15-30 L of solution	Glass, 5 gal.
Type of Dilution System Must provide reproducible supply of toxicant	N/A
Flow Rate Consistent flow rate of 5-10 vol/24 hours, meter systems calibrated before study and checked twice daily during test period	N/A
Biomass Loading Rate Static: ≤ 0.8 g/L at $\leq 17^\circ\text{C}$, ≤ 0.5 g/L at $> 17^\circ\text{C}$; flow-through: ≤ 1 g/L/day	not reported
Photoperiod 16 hours light, 8 hours dark	Not reported
Solvents Not to exceed 0.5 ml/L for static tests or 0.1 ml/L for flow-through tests	No solvents reported

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

Guideline Criteria	Reported Information
<p><u>Nominal Concentrations of Definitive Test</u> Control & 5 treatment levels; dosage should be 60% of the next highest concentration; concentrations should be in a geometric series</p>	<p>Not reported</p>
<p><u>Number of Test Organisms</u> Minimum 10/level, may be divided among containers</p>	<p>not reported</p>
<p><u>Test organisms randomly or impartially assigned to test vessels?</u></p>	<p>Not reported</p>
<p><u>Biological observations made every 24 hours?</u></p>	<p>Yes</p>
<p><u>Water Parameter Measurements</u> 1. <u>Temperature</u> Measured constantly or, if water baths are used, every 6 hrs, may not vary > 1°C 2. <u>DO and pH</u> Measured at beginning of test and every 48 h in the high, medium, and low doses and in the control</p>	<p>Details not reported</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>	<p>Details not reported</p>

12. REPORTED RESULTS

A. General Results

Guideline Criteria	Reported Information
Quality assurance and GLP compliance statements were included in the report?	Yes--exempt from GLP since conducted prior to 10/16/89
Recovery of Chemical	Not reported
Control Mortality Not more than 10% control organisms may die or show abnormal behavior.	No negative controls were used
Raw data included?	No
Signs of toxicity (if any) were described?	Yes; fish became dark and lethargic, lost equilibrium, and expired

Other Significant Results:

B. Statistical Results

Method: Details of analysis were not reported.

96-hr LC₅₀: 3.13 ppm ai 95% C.I.: ~~20.0~~^{2.02} - 4.84 ppm ai

Probit Slope: Not reported NOAEC: 1.2 ppm ai

13. VERIFICATION OF STATISTICAL RESULTS

Due to lack of information, no verification was possible.

14. REVIEWER'S COMMENTS: No comments.

DP Barcode: D260071

MRID # 444349-13

Review of Published Scientific Article

Chemical: Alkyl (C10-16) dimethylamineoxide

PC Code: 000439

Product/Reg. # 0003573-AN Cleaning Care II

Registrant: The Proctor and Gamble Company

Article was: submitted by registrant (MRID # 444349-13)

selected by reviewer during a literature search

Purpose: fulfillment of Guideline requirement (Guideline # 72-2/850.1010)

Was Guideline fulfilled by this study? NO - Supplemental

supplementary information

Reviewed by: Kathryn V. Montague, Biologist, USEPA/AD/RASSB3

Kathryn V. Montague 12/22/99
KVM 12-27-99
PC Code
01-04-2000

Citation: Author: Maki, A.W.

Year: 1979

Title: Correlations Between *Daphnia magna* and Fathead Minnow (*Pimephales promelas*) Chronic Toxicity Values for Several Classes of Test Substances

Journal/vol/pg: J. Fish Res. Board Can. 36: 411-421

Species: *Daphnia magna*

Age or size at testing: <12 h old

Study Type: Life cycle, with acute endpoints (96-h LC50) measured as well

Length of test: 21 days

Test Conditions: flow-through pH: 7.4 Temperature: 21 ± 1° C

hardness: 120 mg/L CaCO₃ DO: 8.5 -9.5 mg/L Alkalinity: not reported

photoperiod: not reported fed during test? Yes--trout chow and alfalfa

Number of treatment levels: 5 Number of organisms per level: 20

Summary: This study was conducted in order to determine the correlation between chronic endpoints for daphnia and fathead minnow for growth, mortality, and certain reproductive parameters in response to exposure to various chemicals. Chemicals were tested individually--the chemical of interest in this case is amine oxide. Twenty daphnids, randomly distributed over 4 replicates containing 5 daphnids each, were exposed to each of five levels of amine oxide (levels

not given in published article). Mortality was recorded daily for the first 4 days, again on day 7, and daily thereafter. Young produced were counted and discarded daily. Production of young generally began on day 7 or 8. Results were analyzed using a computerized probit analysis program. Survival data were used to determine 96-hour and 21-day LC_{50} values with 95% confidence intervals. Young production, mean brood size, and percentage of days that reproduction occurred was converted to percent effects (untreated controls for each test were considered as 0%), and EC_{50} values were determined for each parameter. All calculations were based on mean measured concentrations of the chemical under consideration. NOAEC values were determined as the highest measured test concentration causing no perceptible effects on all parameters of growth, survival, and reproductive performance compared to controls.

The total number of young produced was the most sensitive parameter examined. Results for amine oxide, in mg/L, were:

96-h LC_{50} (95 % conf. limits)	21-d LC_{50} (95 % conf. limits)	Total Young Production EC_{50} (95 % conf. limits)	Average Brood Size EC_{50} (95 % conf. limits)	Percentage of Days Reproduction Occurred EC_{50} (95 % conf. limits)
1.01 (0.85 - 1.21)	0.96 (0.90 - 1.03)	0.88 (0.77 - 1.04)	1.01 (0.95 - 1.07)	1.04 (0.98 - 1.11)

The overall NOAEC for amine oxide was 0.70 mg/L

Reviewer's Comments: The results of this study were not presented in a format to specifically fulfill EPA Guideline requirements; therefore, specific Guideline deviations could not be determined, as the full details of the study methodology were not presented in the published article. Additionally, results could not be verified, since there were no raw data provided.