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MR 280349

3M

October 26, 2004

8EHQ-1004-15860

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**By Hand Delivery**
Document Processing Center (7407)
Office of Pollution, Prevention and Toxics
U.S. Environmental Protection Agency
1200 Pennsylvania Avenue, N. W.
Washington, DC 20460
Attention: Section 8(e) Coordinator

Re: **TSCA Section 8(e) Submissions**

Dear Sir/Madam:

3M Company ("3M") requests that EPA place the attached studies in the TSCA Section 8(e) docket. We have included a master index for these studies identifying the study title, test substance and CAS number. A Confidential Business Information (CBI) version of this index and the studies also is being submitted today pursuant to EPA procedures. 3M has not provided CBI substantiation with this submission, but would be willing to do so at the Agency's request.

3M has concluded that data in these studies may not be, strictly speaking, "corroborative" of previously reported or published information as defined in EPA's reporting guidance or otherwise potentially may warrant 8(e) submission based on EPA's reporting guidance.

3M appreciates EPA's attention to this matter. Please contact the undersigned if you have any questions or require further information regarding this submission.

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Very truly yours,

Katherine E. Reed (9.7.)

Dr. Katherine E. Reed, Ph.D
Staff Vice President
Environmental Technology and Safety Services
(651) 778-4331
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8EHQ-04-15860

8805000006 6/S

# Master Index to Studies Submitted Under TSCA 8(e) by 3M Company on October 26, 2004
## (Confidential Business Information Redacted)

| Title | Substance Information | CAS Information |
|---|---|---|
| Aquatic Toxicity Data Sheet: 48hr *Daphnia Magna* | 1,4-dioxane; heptadecafluoro-1-octanesulfonic acid; linear n-ethyl perfluorooctanesulfonamide; n-ethylperfluorooctanesulfonamidoethyl alcohol; poly(oxy-1,2-ethanediyl), .alpha.-[2-[ethyl[(heptadecafluorooctyl)sulfonyl]amino]ethyl]-.omega.-hydroxy-; poly(oxy-1,2-ethanediyl), .alpha.-[2-[ethyl[(nonafluorobutyl)sulfonyl]amino]ethyl]-.omega.-hydroxy-; poly(oxy-1,2-ethanediyl), .alpha.-[2-[ethyl[(pentadecafluoroheptyl)sulfonyl]amino]ethyl]-.omega.-hydroxy-; poly(oxy-1,2-ethanediyl), .alpha.-[2-[ethyl[(tridecafluorohexyl)sulfonyl]amino]ethyl]-.omega.-hydroxy-; poly(oxy-1,2-ethanediyl), .alpha.-[2-[ethyl[(undecafluoropentyl)sulfonyl]amino]ethyl]-.omega.-hydroxy-; polyethylene glycol; water | 1,4-dioxane (123-91-1); heptadecafluoro-1-octanesulfonic acid (1763-23-1); linear n-ethyl perfluorooctanesulfonamide (4151-50-2); n-ethylperfluorooctanesulfonamidoethyl alcohol (1691-99-2); poly(oxy-1,2-ethanediyl), .alpha.-[2-[ethyl[(heptadecafluorooctyl)sulfonyl]amino]ethyl]-.omega.-hydroxy- (29117-08-6); poly(oxy-1,2-ethanediyl), .alpha.-[2-[ethyl[(nonafluorobutyl)sulfonyl]amino]ethyl]-.omega.-hydroxy- (68298-79-3); poly(oxy-1,2-ethanediyl), .alpha.-[2-[ethyl[(pentadecafluoroheptyl)sulfonyl]amino]ethyl]-.omega.-hydroxy- (68298-81-7); poly(oxy-1,2-ethanediyl), .alpha.-[2-[ethyl[(tridecafluorohexyl)sulfonyl]amino]ethyl]-.omega.-hydroxy- (56372-23-7); poly(oxy-1,2-ethanediyl), .alpha.-[2-[ethyl[(undecafluoropentyl)sulfonyl]amino]ethyl]-.omega.-hydroxy- (68298-80-6); polyethylene glycol (25322-68-3); water (7732-18-5) |
| Multigeneration Daphnid Life Cycle Test | 1,4-dioxane; heptadecafluoro-1-octanesulfonic acid; linear n-ethyl perfluorooctanesulfonamide; n-ethylperfluorooctanesulfonamidoethyl alcohol; poly(oxy-1,2-ethanediyl), .alpha.-[2-[ethyl[(heptadecafluorooctyl)sulfonyl]amino]ethyl]-.omega.-hydroxy-; poly(oxy-1,2-ethanediyl), .alpha.-[2-[ethyl[(nonafluorobutyl)sulfonyl]amino]ethyl]-.omega.-hydroxy-; poly(oxy-1,2-ethanediyl), .alpha.-[2-[ethyl[(pentadecafluoroheptyl)sulfonyl]amino]ethyl]-.omega.-hydroxy-; poly(oxy-1,2-ethanediyl), .alpha.-[2-[ethyl[(tridecafluorohexyl)sulfonyl]amino]ethyl]-.omega.-hydroxy-; poly(oxy-1,2-ethanediyl), .alpha.-[2-[ethyl[(undecafluoropentyl)sulfonyl]amino]ethyl]-.omega.-hydroxy-; polyethylene glycol; water | 1,4-dioxane (123-91-1); heptadecafluoro-1-octanesulfonic acid (1763-23-1); linear n-ethyl perfluorooctanesulfonamide (4151-50-2); n-ethylperfluorooctanesulfonamidoethyl alcohol (1691-99-2); poly(oxy-1,2-ethanediyl), .alpha.-[2-[ethyl[(heptadecafluorooctyl)sulfonyl]amino]ethyl]-.omega.-hydroxy- (29117-08-6); poly(oxy-1,2-ethanediyl), .alpha.-[2-[ethyl[(nonafluorobutyl)sulfonyl]amino]ethyl]-.omega.-hydroxy- (68298-79-3); poly(oxy-1,2-ethanediyl), .alpha.-[2-[ethyl[(pentadecafluoroheptyl)sulfonyl]amino]ethyl]-.omega.-hydroxy- (68298-81-7); poly(oxy-1,2-ethanediyl), .alpha.-[2-[ethyl[(tridecafluorohexyl)sulfonyl]amino]ethyl]-.omega.-hydroxy- (56372-23-7); poly(oxy-1,2-ethanediyl), .alpha.-[2-[ethyl[(undecafluoropentyl)sulfonyl]amino]ethyl]-.omega.-hydroxy- (68298-80-6); polyethylene glycol (25322-68-3); water (7732-18-5) |
| Aquatic Invertebrate Testing - Alkyltins LR 8024-1 | Alkyltins: dibutyltin laurate and dibutyltin-di(2 ethylhexoate) | Dibutyltin laurate (CAS 77-58-7); Dibutyltin-di(2 ethylhexoate) (CAS 2781-10-4) |
| Aquatic Invertebrate Testing - Decosheen Material (LR-8052) | Decosheen Ribbon Materials and pigments: Decosheen Blue in Green Ceres Blue ZV; Decosheen Gold Paste Pigment; Decosheen Royal Blue, Solvent Blue | Decosheen Blue in Green Ceres Blue ZV (CAS 61814-09-3); Decosheen Royal Blue, Solvent Blue (CAS [illegible]); Decosheen Gold Paste Pigment (CAS Number [illegible]) |
| R Scratch Remover (Fathead Minnow) | 55-65% Water; 20-30% Stoddard Solvent; 1-5% Sodium Silicate; 1-5% Potassium Hydroxide; 0.1-3% Nonylphenoxypoly(oxyethylene)ethanol | Water (CAS 7732-18-5); Stoddard Solvent (CAS 8052-41-3); Sodium Silicate (CAS 1344-09-8); Potassium Hydroxide (CAS 1310-58-3); Nonylphenoxypoly(oxyethylene)ethanol (CAS 9016-45-9) |
| S Scratch Remover (Fathead Minnow) | 60-70% Water; 20-30% Stoddard Solvent; 1-5% Sodium Silicate; 0.1-3% Turgitol NP-33 | Water (CAS 7732-18-5); Stoddard Solvent (CAS 8052-41-3); Sodium Silicate (CAS 1344-09-8); Turgitol NP-33 (CAS 9016-45-9) |
| Octanol Water Partition Coefficient | N-methylperfluorooctane sulfonamidoethanol | CAS 24448-09-7 |

# Master Index to Studies Submitted Under TSCA 8(e) by 3M Company on October 26, 2004
## (Confidential Business Information Redacted)

| Title | Substance Information | CAS Information |
|---|---|---|
| CoCl2.6H2O as Co2+ Toxicity to Microtox Reagent | Cobalt (as Co2+ ion) (CoCl2.6H2O) | CAS 7791-13-1 |
| Activated Sludge Respiration Inhibition Test on CoCl2.6H2O as Co ion | Cobalt (as Co2+ ion) (CoCl2.6H2O) | CAS 7791-13-1 |
| Acute Toxicity of CoCl2.6H20 as Co ion to Daphnia magna under Static Exposure Conditions | Cobalt (as Co2+ ion) (CoCl2.6H2O) | CAS 7791-13-1 |
| Acute Toxicity of CoCl2.6H20 as Co ion to Fathead Minnow under Static Exposure Conditions | Cobalt (as Co2+ ion) (CoCl2.6H2O) | CAS 7791-13-1 |
| Freshwater Algae Growth Inhibition Test | Cobalt (as Co2+ ion) (CoCl2.6H2O) | CAS 7791-13-1 |
| *Daphnia magna* 21-Day Chronic Reproduction Study | N-ethylperfluorooctane sulfonamidoethanol | CAS 1691-99-2 |
| Plant Growth Effects of [ ] | [ ] | [ ] |
| Final Report (*Daphnia* and Microtox) | Monomethyl ether of hydroquinone | CAS 150-76-5 |
| Microtox Test Results | 2 Ethylhexyl Acrylate; Isooctyl Acrylate Monomer; 2-Methylbutyl acrylate; Methyl isoamyl acrylate; Isooctyl Acrylate | 2 Ethylhexyl Acrylate (CAS 103-11-7); Isooctyl Acrylate Monomer (CAS 29590-42-9) 2-Methylbutyl acrylate (CAS 44914-03-6); Methyl isoamyl acrylate (CAS 18993-92-1); Isooctyl Acrylate (CAS 29590-42-9) |
| Phytotoxicity Test Results | [ ] | [  ] |

# Master Index to Studies Submitted Under TSCA 8(e) by 3M Company on October 26, 2004
## (Confidential Business Information Redacted)

| Title | Substance Information | CAS Information |
|---|---|---|
| Plant Toxicity Comparison, Young Seedling Growth | [ ] | [ ] |
| *Ceriodaphnia dubia* Survival and Reproduction exposed to Opequon Creek Water Spiked with BETZ 1110 Polymer (November 4, 1987 sample) for seven days under static renewal conditions | BETZ 1110: Non-3M Product - Chemical composition not provided to 3M by manufacturer | MSDS provided by manufacturer states product is "not hazardous" and not "considered to be a carcinogen" |
| *Ceriodaphnia dubia* Survival and Reproduction exposed to Opequon Creek Water Spiked with Betz 1138 Polymer (November 4, 1987 sample) for seven days under static renewal conditions | BETZ 1138: Non-3M Product - Chemical composition not provided to 3M by manufacturer | MSDS provided by manufacturer states product is "not hazardous" and not "considered to be a carcinogen" |
| Toxicity of 1,6 - Hexanediol Diacrylate to *Daphnia magna* | 1,6 Hexanediol diacrylate | CAS 13048-33-4 |
| *Daphnia magna* Chronic Bioassay Under Static Renewal Conditions | Methyl isoamyl acrylate | CAS 18993-92-1 |
| Estimating the Chronic Toxicity of Nalclear 7177 to *Ceriodaphnia* Survival and Reproduction Using Short-Term Tests | Nalclear 7177 wastewater treatment acrylamide/acrylate polymer - Chemical composition not provided to 3M by manufacturer | CAS Information not provided to 3M by manufacturer |
| Acute Toxicity of Isooctyl Acrylate to *Daphnia magna* | Isooctyl Acrylate Monomer | CAS 29590-42-9 |
| Static Acute Toxicity of [ ] to the Daphnid, *Daphnia magna* | Tolyltriazole | CAS 29385-43-1 |
| Static Acute Toxicity of [ ] to the Alga, *Selenastrum capricornutum* | Tolyltriazole | CAS 29385-43-1 |
| Static Acute Toxicity of [ ] to the Daphnid, *Daphnia magna* | [ ] | [ ] |
| Static Acute Toxicity of [ ] to the Fathead Minnow, *Pimephales promelas* | [ ] | [ ] |
| Static Acute Toxicity of [ ] to the Daphnid, *Daphnia magna* | water; propylene-tetrafluoroethylene polymer; tert-butyl alcohol | water (7732-18-5); propylene-tetrafluoroethylene polymer (27029-05-6); tert-butyl alcohol (75-65-0) |

# Master Index to Studies Submitted Under TSCA 8(e) by 3M Company on October 26, 2004
## (Confidential Business Information Redacted)

| Title | Substance Information | CAS Information |
|---|---|---|
| Isooctyl acrylate: Fish, Acute Toxicity Test | Isooctyl Acrylate Monomer | CAS 29590-42-9 |
| Isooctyl Acrylate: *Daphnia sp.* Acute Immobilization Test | Isooctyl Acrylate Monomer | CAS 29590-42-9 |
| Isooctyl Acrylate: Alga, Growth Inhibition Test | Isooctyl Acrylate Monomer | CAS 29590-42-9 |
| Isooctyl Acrylate: *Daphnia sp.* Reproduction Test | Isooctyl Acrylate Monomer | CAS 29590-42-9 |
| Acute Toxicity of [ ] to the mysid, *Mysidopsis bahia* | [ ] | [ ] |
| Final Report (Microtox) | [ ] | [ ] |
| Determination of the Partition Coefficient (N-Octanol/Water) of T-5896 by High Performance Liquid Chromatography (HPLC) | N-methyl perfluorooctane sulfonamido ethanol; N-methyl perfluorooctane sulfonamidethyl acrylate | N-methyl perfluorooctane sulfonamido ethanol (CAS 25268-77-3); N-methyl perfluorooctane sulfonamidethyl acrylate (CAS 24448-09-7) |
| OECD Activated Sludge Respiration Inhibition Test Results | N-Dodecyltrimethylammonium chloride | CAS = 112-00-5 |
| Final Report (Fish Acute Toxicity) | Mirataine CB (30% Cocamidopropyl betaine = Amides, coco, N-(3-(dimethylamino)propyl), alkylation products with chloroacetic acid, sodium salts, 70% Water and Inerts); Mirataine COB (30% Coco/Oleamidopropyl Betaine = 1-Propanaminium, 3-amino-N-(carboxymethyl)-N,N-dimethyl-, N-coco acyl derivs., inner salt) | Cocamidopropyl betaine (CAS 70851-07-9); Coco/Oleamidopropyl Betaine (CAS 61789-40-0) |
| A Flow-Through Life-Cycle Toxicity Test With the Saltwater Mysid (*Mysidopsis bahia*) | Perfluorooctane sulfonate | CAS 1763-23-1 |
| Lithium: Alga, Acute toxicity Tests | Lithium Chloride | CAS 7447-41-8 |
| An Early Life-Stage Toxicity Test With the Fathead Minnow (*Pimephales promelas*) | Perfluorooctane sulfonate | CAS 1763-23-1 |
| Lithium: Fish, Acute toxicity Tests | Lithium Chloride | CAS 7447-41-8 |
| Lithium: *Daphnia*, Acute toxicity Tests | Lithium Chloride | CAS 7447-41-8 |
| Summary of Toxicity Testing on OSCI and OSF | Octane sulfonyl chloride and Octane sulfonyl fluoride | Octane sulfonyl fluoride (CAS 7795-95-1), Octane sulfonyl chloride (CAS 4063-63-5) |
| Toxicity to Microtox Test | Lauryldimethylamineoxide | CAS 1643-20-5 |

# Master Index to Studies Submitted Under TSCA 8(e) by 3M Company on October 26, 2004
## (Confidential Business Information Redacted)

| Title | Substance Information | CAS Information |
|---|---|---|
| Ecotoxicological Testing of CoCl2.6H2O as Co2+ ion (Seed Germination and Root Elongation) | Cobalt (as Co2+ ion) (CoCl2.6H2O) | CAS 7791-13-1 |

Form 6747 11-8

# TECHNICAL REPORT SUMMARY

Date 7/17/81

TO: TECHNICAL COMMUNICATIONS CENTER – 201-2CN

*(Important – If report is printed on both sides of paper, send two copies to TCC.)*

Division
Environmental Engineering and Pollution Control

Project
New Method Development

Report Title
Multigeneration Daphnid Life Cycle Test (Technology Transfer)

No. of Pages Including Coversheet

SECURITY ▶ ☐ Open (Company Confidential) ☒ Closed (Special Authorization)

3M CHEMICAL REGISTRY ▶ New Chemicals Reported ☐ Yes ☒ No

KEYWORDS: (Select terms from 3M Thesaurus. Suggest other applicable terms.)

Daphnids
Life-cycle
Multi-generation
Reproduction
Survival
Toxicant
Environmental
Laboratory

CURRENT OBJECTIVE:

To determine the effect of repeated exposures to a toxicant on daphnids (Technology Transfer).

REPORT ABSTRACT: (200-250 words) This abstract information is distributed by the Technical Communications Center to alert 3M'ers to Company R&D. It is Company confidential material.

Information Liaison Initials:

CONFIDENTIAL BUSINESS INFORMATION
SUBJECT TO PROTECTION UNDER THE
TOXIC SUBSTANCES CONTROL ACT
AND OTHER LAWS HAS BEEN
REDACTED FROM THIS DOCUMENT

**3M CONFIDENTIAL**

## Introduction

Chronic life-cycle testing procedures are utilized to provide information relative to the long term exposure effects of a possible toxicant on a test organism. To illustrate the universality of this concept, standardized chronic life-cycle protocols are being developed in the United States by both the American Society of Testing Materials (1979) and the United States Environmental Protection Agency (1980). The Organization of Economic Cooperation and Development (24 member countries) has included a Daphnia reproductive study as a data component for the development of a minimum premarket data set for new chemicals prior to entering the market place (1980).

Daphnids are a representative invertebrate species and reportedly are generally more sensitive to toxicants vis a vis either freshwater algae and/or fish. Additionally life-cycle studies performed using Daphnia magna are customarily completed within 28 days as opposed to fish life-cycle studies which require nine months. Similarly, the former tests are less expensive to perform, utilizing less equipment and are less labor intensive. Most importantly the daphnid studies are relatively simple to perform. Results of daphnid life-cycle studies are also useful in making hazard evaluations and have been utilized in deriving water quality data (FR 44, 1979).

Because of the aforementioned usefulness of a life-cycle test and more specifically a daphnid life-cycle test a technology transfer program was initiated so that these tests could be performed on a routine basis in the Environmental Laboratory facility. It is the purpose of this report to outline problems encountered during this period and to list the results obtained while performing a multigeneration daphnid life-cycle test.

## Methods

The test protocol utilized for this study was modeled after that described by the ASTM (1979).

Daphnia magna (water fleas) were obtained from Mogul Education(a) and the culture was maintained in an 18-liter glass chamber filled with carbon-filtered well water. Monitoring equipment used included ASTM thermometers, Orion temperature-compensating pH meter and a Yellow Springs dissolved oxygen (DO) meter. Temperature was maintained at 20±2°C and at a pH of 7.0. Organisms were fed three times per week using a 4:1 mixture of Tetra Min(b) and active dry yeast, concentration of 30 mg/l.

(a)Somerset, Wisconsin.
(b)Commercial fish food of known composition.

Experiments were conducted using a Sherer Growth Chamber equipped with a temperature recorder and automatic circuitry for maintaining the temperature within +1°C of the experimental temperature. A daily photoperiod of 16 hours light and 8 hours dark with a 30-minute transition period was maintained throughout the test period.

To initiate a test, adult daphnids were segregated for the purpose of obtaining first instars. These first instars were distributed randomly to beakers containing 200 ml well water and varying concentrations of [ ] cc 7718-18 e g; 0, 65, 100, 150, 240 and 370 ug/l. A total of 154 first instar daphnids were required for each test. A schema for a single generation test is appended to this report. The mid-concentration, 150 ug/l, was selected based on a calculated acute static daphnid $LC_{50}$ value. This concentration was approximately 1/10th of the $LC_{50}$ value. Following completion of the multigeneration test the $LC_{50}$ for daphnids was repeated using the ASTM protocol (1980). Fresh stock solutions of [ ] having a concentration of 100 ug/l were prepared thrice weekly. At these solution renewal periods food was added to aerated well water, pH, temperature and dissolved oxygen were also measured. To supplement these determinations, water quality analyses are routinely performed on a monthly basis. Results of these tests performed during the multigeneration test form Appendix B.

At the time of solution renewal the following observations were made: number surviving, number of young, behavioral effects. Upon termination of the test, length of the surviving daphnids was also recorded.

Statistical treatment of the data included calculations of mean values + standard deviations, analysis of variance. Thomas Brackley of IS & DP assisted in the statistical handling of the data generated by this study.

## Results and Discussion

Among the parameters selected to evaluate possible toxicant effects of either continuous or intermittant exposure to [ ] was the survival time. A tabulation of this parameter, Table 1, indicates that when continuously exposed to [ ] daphnids may readily adapt to the presence of this toxicant. Thus from $F_1$-$F_3$ the percent survival increased at each generation. This postulate would of necessity require substantiation based on repetitive testing under identical conditions.

In the situation where the $F_2$ generation was not exposed to [ ] but $F_3$ was reexposed to this chemical, highy variable results were noted. One may conjecture as to the significance of these data; e g; $F_1$ and $F_2$ daphnids were similarly stressed, that is $F_1$ was stressed by the presence of chemical while $F_2$ was stressed by its absence and the $F_3$ result, 70% survival, may be attributable to information transfer from $F_1$ to $F_3$ inter. these are highly speculative interpretations.

In an unreported study, we did find 73% daphnid survival beyond 28 days in a reproductive study (1981).

Based on the foregoing and a discussion of daphnid survival variability by Muller (1980) and Parkhurst et al (1981) the more tenable conclusion to be derived from the data presented as Table 1, is a simple confirmation of biological variability due to use of living organisms.

Table 1 Survival Percentage of Daphnids Used in Multigeneration Study Independent of [ ] Exposure

| Generation | Percent Survival |
|---|---|
| $F_1$ | 35 |
| $F_2$ Continuous[a] | 63 |
| $F_2$ Intermittant[b] | 32 |
| $F_3$ Continuous | 84 |
| $F_3$ Intermittant | 70 |

a Continuous refers to nature of toxicant exposure.

b Intermittant refers to fact that $F_2$ generation was not exposed to toxicant. Reexposure did occur at $F_3$ and has been labelled $F_3$ intermittant.

Table 2 illustrates the variability in time to initial brood release by daphnids, a factor shown to be independent of the presence or absence of toxicant. The $F_2$ generation showed a faster initial brood release time. This result coincides with those obtained recently in our unpublished study wherein all initial broods were released from day 6 to 10. The former value was most prevalent while the 10-day period for release was an exception. Again this finding is not novel as Parkhurst et al (1981) reported a similar occurrence. The variability noted by the aforementioned authors is much greater than those presently being reported. The [ ] variability was between generations, hence not randomized. Again it is recommended that additional studies be undertaken to demonstrate the presence or absence of reproducability of this parameter.

Table 2 Time to first brood release of Daphnia magna prior to and during exposure to [ ]

| Conc. ug/l | $F_1$ Mean | ±S.D. | $F_2$[a] Mean | ±S.D. | $F_2$[a] Inter Mean | ±S.D. | $F_3$[b] Mean | ±S.D. | $F_3$[c] Inter Mean | ±S.D. |
|---|---|---|---|---|---|---|---|---|---|---|
| Cont. | 11.8 | 0.6 | 7.0 | 1.1 | 7.5 | 1.4 | 10.9 | 1.7 | 13.3 | 0.8 |
| 65 | 10.8 | 0.7 | 6.5 | 0.7 | — | | 11.1 | 3.0 | 12.0 | 1.7 |
| 100 | 10.9 | 1.5 | 6.0 | 0 | — | | 12.0 | 0 | 12.0 | 1.7 |
| 150 | 10.9 | 1.0 | 7.8 | 0.7 | — | | 12.0 | 0 | 12.0 | 0 |
| 240 | 10.4 | 0.9 | 7.3 | 0.6 | — | | 13.7 | 3.7 | 12.0 | 1.6 |
| 370 | 11.5 | 2.1 | 7.8 | 0.7 | — | | 12.2 | 1.3 | 12.0 | 1.6 |

a Values expressed as days.

b Continuous exposure to toxicant $F_1$-$F_3$.

c Intermittant-$F_2$ inter was not exposed to toxicant. $F_3$ inter was reexposed to toxicant.

Table 3 lists the effect of [ ] exposure on brood production of Daphnia magna. Because of variability in responsiveness no statistically significant trends were apparent. In no phase of this study was a toxicant effect on any of the reproductive parameters apparent.

Parkhurst et al (1981) indicated that the average number of young produced per female was found to be a sensitive and most reliable indicator for estimating the "lowest rejected concentration tested." Table 4 depicts the results obtained when the collected data from this multigeneration study was tabulated. These data show similar trends to those depicted as Table 3. Because of data variability no specific toxicant effect could be determined.

To determine whether the doses used in this multigeneration study were consistent with acute static daphnid $LC_{50}$ studies previously performed and were equally applicable to $LC_{50}$ data from the identical [ ] sample having a shelf-life of several years these studies were repeated. The results of this study are shown as an aquatic toxicity data sheet. The composite $LC_{50}$ values for the two studies performed was 1 mg/l with 95% confidence limits of 0.8-1.2 mg/l. Since life-cycle study protocols require that the mid-concentration be equivalent or approximates 1/10th of the $LC_{50}$, our data do suggest that the values were indeed quite similar.

Table 3 Effect of 28-day Exposure to [ ] on brood production of Daphnia magna[c]

| Conc. ug/l | $F_1$ Mean | +S.D. | $F_2$[a] Mean | +S.D. | $F_2$[b] Inter Mean | +S.D. | $F_3$[a] Mean | +S.D. | $F_3$[a] Inter Mean | +S.D. |
|---|---|---|---|---|---|---|---|---|---|---|
| Cont. | 4.8 | 2.0 | 5.5 | 1.8 | 4.6 | 2.2 | 4.4 | 1.6 | 4.3 | 0.6 |
| 65 | 3.8 | 2.1 | 7.2 | 2.6 | — | | 4.6 | 0.2 | 5.7 | 0.6 |
| 100 | 3.7 | 2.4 | 7.1 | 1.1 | — | | 4.8 | 1.6 | 4.3 | 1.2 |
| 150 | 1.3 | 0.6 | 6.2 | 1.6 | — | | 4.3 | 2.0 | 5.0 | 0.6 |
| 240 | 3.3 | 2.4 | 4.6 | 2.1 | — | | 4.9 | 1.1 | 4.3 | 1.0 |
| 370 | 2.2 | 2.1 | 5.1 | 2.2 | — | | 5.0 | 0.8 | 5.0 | 1.5 |

a Continuous exposure to toxicant $F_1$-$F_3$.

b Intermittant-$F_2$ inter was not exposed to toxicant. $F_3$ inter was reexposed to toxicant.

c Values expressed as number of broods.

Table 4 Average number of young produced by a female daphnid exposed to [ ] for 28 days[a]

| Conc. ug/l | $F_1$ Mean | +S.D. | $F_2$[b] Mean | +S.D. | $F_2$[c] Inter Mean | +S.D. | $F_3$[b] Mean | +S.D. | $F_3$[c] Inter Mean | +S.D. |
|---|---|---|---|---|---|---|---|---|---|---|
| Cont. | 19.9 | 6.3 | 19.0 | 4.9 | 22.3 | 9.2 | 20.9 | 5.7 | 15.9 | 8.6 |
| 65 | 22.8 | 7.4 | 25.0 | 3.7 | — | | 17.8 | 4.2 | 21.3 | 8.2 |
| 100 | 18.1 | 12.0 | 20.9 | 4.5 | — | | 19.2 | 6.5 | 16.3 | 3.5 |
| 150 | 9.2 | 4.1 | 20.4 | 2.6 | — | | 17.7 | 8.0 | 18.6 | 4.0 |
| 240 | 13.7 | 12.5 | 15.6 | 6.8 | — | | 22.8 | 4.7 | 21.2 | 4.3 |
| 370 | 8.6 | 12.0 | 15.6 | 6.4 | — | | 21.1 | 3.6 | 17.6. | 1.2 |

a Values expressed as number of young per female.

b Continuous exposure to toxicant $F_1$-$F_3$.

c Intermittant-$F_2$ inter no exposure to toxicant, $F_3$ inter reexposed to toxicant.

## Conclusions

In this technology transfer exercise it was demonstrated that biological variability could account for many of the results obtained. This series of experiments again provides further evidence that caution be used in the interpretation of isolated results and the absolute necessity for increasing the number of studies performed.

## References

American Society for Testing and Materials: Proposed Standard Practice for Conducting Renewal Life Cycle Toxicity Tests with Daphnids, Draft No. 5. American Society for Testing and Materials. Committee E 35.21, Philadelphia, Pennsylvania (1979).

United States Environmental Protection Agency: Chronic Toxicity Test Standard Using Daphnids in Renewal and Flow-Through Systems, Draft. Environmental Protection Agency, Washington, D.C. (1980).

Chemical Regulation Reporter: Data Components for the OECD Minimum Premarketing Set of Data, p. 786, September 12, 1980.

United States Environmental Protection Agency: Water Quality Criteria, Federal Register 44: 15926-15981, March 15, 1979.

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