ROBUST SUMMARY FOR TRIISOPROPYLBORATE

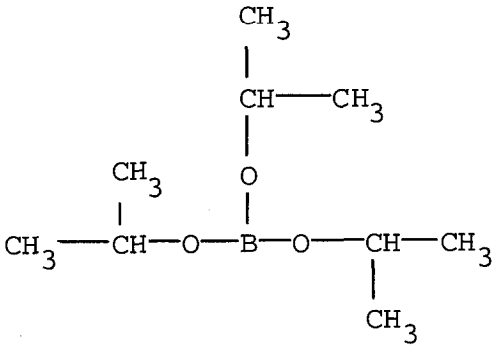
Existing published and unpublished data were collected and scientifically evaluated to determine the best possible study or studies to be summarized for each required endpoint. In the spirit of this voluntary program, other data of equal or lesser quality are not summarized, but are listed as related references at the end of each appropriate section, with a statement to reflect the reason why these studies were not summarized.

1.0 Substance Information

CAS Number: 5419-55-6

Chemical Name: Boric acid (H_3BO_3), tris(1-methylethyl)ester

Structural Formula:



Other Names:
Triisopropylborate (TIPB)
Boric acid, triisopropyl ester
Boric acid (H_3BO_3), triisopropyl ester
Boron isopropoxide
Boron triisopropoxide
Isopropyl borate
Triisopropoxy borane
Triisopropoxy boron
Triisopropyl orthoborate
Trisisopropxyborane

Exposure Limits: No Data

2.0 Physical/Chemical Properties

2.1 Melting Point

Value:	-59°C
Decomposition:	No Data
Sublimation:	No Data

Pressure:	No Data
Method:	No Data
GLP:	Unknown
Reference:	Lewis, R. J., Sr. (2000). Sax's Dangerous Properties of Industrial Materials, 10th ed., p. 2157, John Wiley & Sons, Inc., New York.
Reliability:	Not assignable because limited study information was available.

Additional Reference for Melting Point:

DuPont Co. (2003). DuPont Material Safety Data Sheet 6332CR (September 4).

2.2 Boiling Point

Value:	140°C
Decomposition:	No Data
Pressure:	760 mm Hg
Method:	No Data
GLP:	Unknown
Reference:	Lide, D. R. (ed.) (2001-2002). Handbook of Chemistry and Physics, 82nd ed., CRC Press, Boca Raton, FL.
Reliability:	Not assignable because limited study information was available.

Additional References for Boiling Point:

DuPont Co. (2003). DuPont Material Safety Data Sheet 6332CR (September 4).

Lewis, R. J., Sr. (2000). Sax's Dangerous Properties of Industrial Materials, 10th ed., p. 2157, John Wiley & Sons, Inc., New York.

2.3 Density

Value:	0.8251 g/cm^3
Temperature:	20°C
Method:	No Data
GLP:	Unknown
Results:	No additional data.
Reference:	Lide, D. R. (ed.) (2001-2002). Handbook of Chemistry and Physics, 82nd ed., CRC Press, Boca Raton, FL.
Reliability:	Not assignable because limited study information was available.

Additional Reference for Density:

Lewis, R. J., Sr. (2000). Sax's Dangerous Properties of Industrial Materials, 10th ed., p. 2157, John Wiley & Sons, Inc., New York.

2.4 Vapor Pressure

Value:	100 mm Hg
Temperature:	25.5°C
Decomposition:	No Data
Method:	No Data
GLP:	Unknown
Reference:	DuPont Co. (2003). DuPont Material Safety Data Sheet 6332CR (September 4).
Reliability:	Not assignable because limited study information was available.

Additional References for Vapor Pressure: None Found.

2.5 Partition Coefficient (log Kow)

Value:	Triisopropylborate: 0.83 (estimated) Isopropanol (hydrolysis product) = 0.28 (estimated) Boric acid (hydrolysis product) = -0.22 (estimated)
Temperature:	25°C
Method:	Modeled. KOWWIN, v.1.67, module of EPIWIN v.3.11 (Syracuse Research Corporation). KOWWIN uses "fragment constant" methodologies to predict log P. In a "fragment constant" method, a structure is divided into fragments (atom or larger functional groups) and coefficient values of each fragment or group are summed together to yield the log P estimate.
GLP:	Not Applicable

Reference:	Meylan, W. M. and P. H. Howard (1995). J. Pharm. Sci., 84:83-92.
Reliability:	Estimated value based on accepted model.

Additional References for Partition Coefficient (log Kow): None Found.

2.6 Water Solubility

Value:	0% (hydrolyzes)
Temperature:	No Data
pH/pKa:	No Data
Method:	No Data
GLP:	Unknown
Reference:	DuPont Co. (2003). DuPont Material Safety Data Sheet 6332CR (September 4).
Reliability:	Not assignable because limited study information was available.

Additional References for Water Solubility: None Found.

2.7 Flash Point

Value:	27.8°C
Method:	TCC
GLP:	Unknown
Reference:	Lewis, R. J., Sr. (2000). Sax's Dangerous Properties of Industrial Materials, 10th ed., p. 2157, John Wiley & Sons, Inc., New York.
Reliability:	Not assignable because limited study information was available.

Additional References for Flash Point:

DuPont Co. (2003). DuPont Material Safety Data Sheet 6332CR (September 4).

Lide, D. R. (ed.) (2001-2002). Handbook of Chemistry and Physics, 82nd ed., CRC Press, Boca Raton, FL.

2.8 Flammability

Results:	Flammable liquid
Method:	No Data
GLP:	Unknown
Reference:	DuPont Co. (2003). DuPont Material Safety Data Sheet 6332CR (September 4).
Reliability:	Not assignable because limited study information was

available.

Additional References for Flammability: None Found.

3.0 Environmental Fate

3.1 Photodegradation

Concentration:	No Data
Temperature:	No Data
Direct Photolysis:	The hydrolysis products, isopropanol and boric acid, are not expected to be subject to direct photolysis (Harris, 1990), due to a lack of significant absorbtivity above 290 nm.
Indirect Photolysis:	Triisopropylborate has an estimated half-life due to OH radical oxidation of 10.55 hours.
Breakdown Products:	No Data
Method:	Direct Photolysis: Inspection of chemical structure. Indirect Photolysis: Modeled
GLP:	Not Applicable
Reference:	AOPWIN, v1.91 module of EPIWIN v.3.11. Meylan, W. M. and P. H. Howard (1993). Chemosphere, 26:2293-99. The model used assumptions of a 24-hour day and an ambient hydroxyl radical concentration of $0.5x10^6$ molecules/cm^3. Harris, J. C. (1990). Rate of Aqueous Photolysis, Chapter 8, In: Handbook of Chemical Property Estimation Methods, Lyman, W. J. et al. (eds.), American Chemical Society, Washington, DC.
Reliability:	Estimate based on known qualitative structure-activity relationships.

Additional References for Photodegradation: None Found.

3.2 Stability in Water

Concentration:	No Data
Half-life:	Unstable in water
% Hydrolyzed:	No Data
Method:	No Data
GLP:	Not Applicable
Reference:	DuPont Co. (2003). DuPont Material Safety Data Sheet 6332CR (September 4).
Reliability:	Not assignable because limited study information was available.

Additional References for Stability in Water: None Found.

3.3 Transport (Fugacity)

Media: Air, Water, Soil, and Sediments

Distributions:

Compartment	% of total distribution	½ life (hours) (advection + reaction)
Air	7.93	10.5
Water	61.5	360*
Soil	30.5	720
Sediment	0.12	3240

* ½ life will be lowered by hydrolysis, which is not treated by this model

Adsorption Coefficient: Koc = 2.77

Desorption: No Data

Volatility: Henry's Law Constant = 0.000647 atm-m^3/mole

Method: Modeled assuming equal emission to air, water, and soil of triisopropylborate (CAS Registry # 5419-55-6).

Henry's Law Constant: 0.000647 atm-m^3/mole (calc VP/Wsol)
Vapor Pressure: 100 mm Hg (user-entered)
Log Kow: 0.83 (KOWWIN program)
Soil Koc: 2.77 (calc by model)

Henry's Law Constant - HENRYWIN v.3.10 module of EPIWIN v.3.11 (Syracuse Research Corporation). Henry's Law Constant (HLC) is estimated by two separate methods that yield two separate estimates. The first method is the bond contribution method and the second is the group contribution method. The bond contribution method is able to estimate many more types of structures; however, the group method estimate is usually preferred (but not always) when all fragment values are available.

Koc – Calculated from log Kow by the Mackay Level III fugacity model incorporated into EPIWIN v.3.11 (Syracuse Research Corporation).

Environmental Distribution - Mackay Level III fugacity model, in EPIWIN v.3.11 (Syracuse Research Corporation). Emissions (1000 kg/hr) to air, water, and soil compartments.

GLP: Not Applicable

Reference: HENRYWIN –
J. Hine and P. K. Mookerjee (1975). J. Org. Chem.,

40(3):292-298.

Meylan, W. and P. H. Howard (1991). Environ. Toxicol. Chem., 10:1283-1293.

Fugacity - The methodology and programming for the Level III fugacity model incorporated into EPIWIN v.3.11 (Syracuse Research Corporation) were developed by Dr. Donald MacKay and coworkers and are detailed in:

Mackay, D. (1991). Multimedia Environmental Models: The Fugacity Approach, pp. 67-183, Lewis Publishers, CRC Press.

Mackay, D. et al. (1996). Environ. Toxicol. Chem., 15(9):1618-1626.

Mackay, D. et al. (1996). Environ. Toxicol. Chem., 15(9):1627-1637.

Reliability: Estimated values based on accepted models.

Additional References for Transport (Fugacity): None Found.

3.4 Biodegradation

No data for triisopropylborate exists, since the chemical rapidly hydrolyzes to boric acid and isopropanol. Therefore, for this robust summary, supporting data for the hydrolysis products are presented.

Data for Hydrolysis Product: Isopropanol

Value:	The hydrolysis product, isopropanol, is readily biodegradable, and averaged 86% of ThBOD.
Breakdown Products:	No Data
Method:	MITI 14-day test (OECD Guideline 301C).
GLP:	Yes
Reference:	Chemicals Evaluation and Research Institute, Japan http://qsar.cerij.or.jp/cgi-bin/QSAR/index.cgi?e
Reliability:	High because a scientifically defensible or guideline method was used.

Data for Hydrolysis Product: Boric Acid

Value:	The hydrolysis product, boric acid, is not subject to biodegradation.
Breakdown Products:	Not Applicable

Method:	Not Applicable
GLP:	Not Applicable
Reference:	Not Applicable
Reliability:	Not Applicable

Additional References for Biodegradation: None Found.

3.5 Bioconcentration

Value:	BCF – Isopropylborate (parent): 0.5 (estimated) BCF - Isopropanol (hydrolysis product): 0.5 (estimated) BCF – Boric acid (hydrolysis product): 0.5 (estimated)
Method:	Modeled. BCFWIN v. 2.15 module of EPINWIN v.3.11 (Syracuse Research Corporation). BCFWIN estimates the bioconcentration factor (BCF) of an organic compound using the compound's log octanol-water partition coefficient (Kow) with correction factors based on molecular fragments.
GLP:	Not Applicable
Reference:	"Improved Method for Estimating Bioconcentration Factor (BCF) from Octanol-Water Partition Coefficient", SRC TR-97-006 (2nd Update), July 22, 1997; prepared for Robert S. Boethling, EPA-OPPT, Washington, DC, Contract No. 68-D5-0012; prepared by William M. Meylan, Philip H. Howard, Dallas Aronson, Heather Printup, and Sybil Gouchie, Syracuse Research Corp.
Reliability:	Estimated values based on accepted model.

Additional References for Bioconcentration: None Found.

4.0 Ecotoxicity

4.1 Acute Toxicity to Fish

No data for triisopropylborate exists, since the chemical rapidly hydrolyzes to boric acid and isopropanol. Therefore, for this robust summary, supporting data for the hydrolysis products are presented.

Data for Hydrolysis Product: Isopropanol

Type:	**96-hour LC_{50}**
Species:	*Pimephales promelas,* fathead minnow
Value:	9640-10,400 mg/L
Method:	No specific test guideline was reported; however, a scientifically defensible approach was used to conduct the study.

The 96-hour LC_{50} was determined with Environmental

Research Laboratory-Duluth cultured fathead minnows using a flow-through proportional diluter and a modified Benoit continuous-flow minidiluter system. Lake Superior water maintained at 25±1°C was used in the test, and data were recorded for hardness, total alkalinity, pH, and dissolved oxygen.

Twenty to 25, 30-day-old fish, each weighing approximately 0.12 g were randomly divided among the test tanks (control and 5 different concentrations in duplicate). Fish were not fed during the test. Deaths were recorded after 1, 3, 6, 12, 24, 48, 72, and 96 hours, and the median lethal concentration (LC_{50}) was computed using the trimmed Spearman-Karber method.

Concentrations of test chemical in water were measured daily at each exposure level. Water was analyzed by gas chromatography or UV spectroscopy and spectrofluorimetry.

GLP: No

Test Substance: Isopropanol, purity not reported

Results: Routine measures of hardness and total alkalinity of test water yielded mean values of 45.5 and 42.2 mg/L as $CaCO_3$, respectively. The mean of the pH was 7.5, and dissolved oxygen was always greater than 60% of saturation.

Reference: Veith, G. D. et al. (1983). Can. J. Fish. Aquatic. Sci., 40:743-748.

Reliability: High because a scientifically defensible or guideline method was used.

Data for Hydrolysis Product: Boric acid

Type: 96-hour LC_{50}

Species: *Oncorhynchus tshawytscha,* Chinook salmon
Oncorhynchus kisutch, Coho salmon

Value: 725 mg/L (95% confidence interval, 590-890 mg/L; Chinook salmon; freshwater)

600 mg/L (95% confidence interval, 511-706 mg/L; Chinook salmon; brackish water)

447 mg/L (95% confidence interval, 356-561 mg/L; Coho salmon; fresh water)

600 mg/L (95% confidence interval, 511-705 mg/L; Coho salmon; brackish water)

Method: Static acute toxicity tests were conducted according to procedures recommended by ASTM (Report E-729).

Fish were fed a commercial salmon diet. Tests were conducted in 3 reconstituted waters. The characteristics of 2 of the dilution waters were designated to simulate water qualities for major anions and cations in which standardized San Luis Drain water without trace elements, was diluted 10-fold in a standardized fresh receiving water or 22.5-fold in a standardized brackish receiving water. The 3rd water was standard soft water prepared as recommended by the U.S. EPA for use in acute toxicity tests with fish. The various water types were reconstituted in reverse osmosis-deionized water by varying the type and quantity of mineral salts added. Water quality characteristics were measured according to methods recommended by the APHA and U.S. EPA. For each type of dilution water, there was only a small amount of variation among batches prepared, as evidenced by the low coefficient of variation (<5%) for each measured characteristic. Test solutions were prepared in deionized water on the day of use or by adding the test substance directly to the test vessel. Nominal concentrations reported were expressed as the total element added as determined from the certificate of analysis for the test substance.

Static 96-hour exposures were conducted in 19.6-L glass jars containing 15 L of test solution and maintained at 12±1°C in temperature-controlled water baths. Each test consisted of exposing groups of 10 fish to a series of toxicant concentrations that differed by 60% between treatments, and a control. For large, advanced fry, duplicate sets of jars were used and only 5 fish were stocked in each jar to maintain loading densities of 0.8 g/L or lower. Observations of mortality and abnormal behavioral responses were made at 24-hour intervals, and all dead fish were removed after each observation.

In 1 set of tests, 2 life stages of Chinook salmon and coho salmon were tested with boric acid. One life stage composed of swim-up fry 8-12 weeks old (post hatch) tested in fresh water. The 2nd life stage composed of advanced fry 15-21 weeks old tested in brackish water. Chinook salmon were also tested in soft water.

In tests conducted in the 2 site-specific waters, pH was measured at the beginning and end of the test.

	Eyed eggs, alveins, and swim-up fry of Chinook salmon were tested in soft water to determine the relative sensitivity of those various life stages to boron. Tests with eyed eggs and alevins were conducted in the dark in glass jars containing test solution. Due to their rapid development, these life stages were acclimated for only 24 hours before testing. Swim-up fry were tested in solution under natural laboratory lighting. Groups of 25 eggs, 10 alevins, or 10 swim-up fry were exposed to a series of toxicant concentrations that differed by 56% between treatments, and a control. Criteria of death for eggs were the presence of an opaque (whitened) membrane. Alevins were examined under 30x magnification for the absence of a heartbeat, which was the criterion for death. The method of Litchfield and Wilcoxon was used to calculate LC_{50} values. The Standard Error of the Difference was used to determine statistical differences in LC_{50} values.
GLP:	No
Test Substance:	Boric acid, reagent grade or highest grade available from supplier
Results:	The 24-hour LC_{50} of Chinook and coho salmon (fresh or brackish water) was >1000 mg/L. Boron was relatively non-toxic. Young coho salmon tested in fresh water were less tolerant than older fish tested in brackish water and Chinook salmon tested in either dilution water. The relative sensitivity of various early life stages of Chinook salmon to boron changed significantly as the fish developed. Fry were consistently more sensitive than either the embryos or alevins to boron. The pH in test concentrations and controls were not markedly different, ranging from 6.5 to 8.1. No significant differences in 96-hour LC_{50} values were found among dilution water qualities, thus indicating that differences in water quality characteristics tested did not modify the toxicity of these chemicals.
Reference:	Hamilton, S. J. and K. J. Buhl (1990). Arch. Environ. Contam. Toxicol., 19:366-373.
Reliability:	Medium because a suboptimal study design (nominal test concentrations) was used.
Type:	**96-hour LC_{50}**
Species:	*Ptychocheilus lucius*, Colorado squawfish *Xyrauchen texanus*, Razorback sucker *Gila elegans*, Bonytail

Value:	279 mg/L, as boron (Colorado squawfish; swimup fry 17-31 day)
	>100 mg/L, as boron (Colorado squawfish; juvenile 99-115 day)
	527 mg/L, as boron (Colorado squawfish; juvenile 193-207 day)
	233 mg/L, as boron (Razorback sucker; swimup fry 10-17 day)
	279 mg/L, as boron (Razorback sucker; juvenile 133-139 day)
	>100 mg/L, as boron (Razorback sucker; juvenile 176-178 day)
	280 mg/L, as boron (Bonytail; swimupfry 11-18 day)
	>100 mg/L, as boron (Bonytail; juvenile 138-145 day)
	553 mg/L, as boron (Bonytail; juvenile 220-234 day)
Method:	Static acute toxicity test procedures closely followed those outlined by the American Society for Testing and Materials (ASTM) (1989).
	Fish were fed a commercial diet supplemented for the first 30 days with live nauplii of brine shrimp. The water quality for egg and larval culture was pH 7.7-7.9, hardness 233-330 mg/L, and alkalinity 174-226 mg/L. All tests were conducted in a reconstituted water quality designed to simulate site-specific conditions for major cations and anions, without trace elements, in the Green river. Reconstituted water was prepared and water quality characteristics were measured for each tank of dilution water.
	Test solutions of boric acid were prepared as a stock solution or by adding the test substance directly to the test vessel. Stock solutions were formulated in deionized water on the day of use. Nominal concentrations reported were expressed as the total inorganic toxicant added, as determined from the certificate of analysis for the test substance.
	Each test consisted of exposing groups of 10 fish to a

geometric series of 6-8 nominal concentrations and a control for 96 hours. Tests with fry were conducted in 3.8 L-glass jars filled with 3 L of test solution and those with juveniles in 19.6-L glass jars filled with 15 L of test solution. For larger juveniles (1.2 g), duplicate sets of jars were used, and only 5 fish were placed in each jar to maintain loading densities of 0.8 mg/L or less. Temperature was maintained at 25±1°C by immersing the jars in temperature-controlled water baths. Because of their rapid rate of development, fry were acclimated to the dilution water for only 24 hours before testing. Juveniles were acclimated simultaneously to the dilution water and test temperature over a 2-day period and were held in the dilution water for 2 days prior to testing. The fish were not fed during acclimation or testing.

At the start of each test, fish were randomly distributed to the test vessels within 30 minutes after the addition of the toxicant. To minimize handling stress, fry were first transferred to 50-mL beakers, containing a small volume of dilution water. After 10 fish were placed in a beaker, most of the water was decanted and the fish were gently poured into the jars. Juveniles were carefully netted from the holding tank and distributed in groups of 2 to each jar. Mortality was recorded, and all dead fish were removed at 24-hour intervals. Fry without perceivable movement of the pectoral fins were pipetted from the jar and examined under 30x magnification for the absence of a heartbeat, which was the criterion for death. Total length and weight of the control fish were measured at the end of the tests.

Dissolved oxygen and pH were measured at the beginning and end of the tests in the control, low, medium, and high treatments with live fish present.

The LC_{50} values were calculated by the binomial method or the moving average-angle method, depending on the number of partial kills in a test, using a computer program. In tests with boron where less than half the fish died in the highest test concentration, the LC_{50} was reported as being greater than that concentration. For tests in which no partial mortalities occurred, the confidence intervals were given as follows: the upper limit was the lowest test concentration with 100% mortality, and the lower limit was the lowest test concentration with 0% mortality. All LC_{50} values were expressed as nominal concentrations of the inorganic toxicant, not the inorganic compound. The standard error of

	the difference was used to determine statistical differences in LC_{50} values.
GLP:	No
Test Substance:	Boric acid, purity not reported
Results:	There was no significant difference among the 3 species in their sensitivity to the test substance. In general, the swimup life stage was more sensitive to boron than the 2 older life stages of the 3 species. There was no mortality in the control treatments from the tests. Dissolved oxygen concentrations were maintained at or greater than 40% saturation in most tests; however, fish in control and low treatment tests with <40% saturation displayed no signs of stress. The pH of the test solutions ranged from 7.0 to 8.5 at 96 hours of exposure.
Reference:	Hamilton, S. J. (1995). Ecotoxicol. Environ. Safety, 30:134-142.
Reliability:	Medium because a suboptimal study design (nominal test concentrations) was used.
Type:	**LC_{50}**
Species:	*Salmo gairdneri*, rainbow trout *Ictalurus punctatus*, channel catfish *Casrassius auratus*, goldfish
Value:	150 ppm (95% confidence interval, 90-249 ppm; rainbow trout; at hatching; soft water) 100 ppm (95% confidence interval, 70-142 ppm; rainbow trout; 4 day post-hatching; soft water) 100 ppm (95% confidence interval, 61-163 ppm; rainbow trout; at hatching; hard water) 79 ppm (95% confidence interval, 35-165 ppm; rainbow trout; 4 days post-hatching; hard water) 220 ppm (95% confidence interval, 167-290 ppm; channel catfish; at hatching; soft water) 155 ppm (95% confidence interval, 111-217 ppm; channel catfish; 4 day post-hatching; soft water) 102 ppm (95% confidence interval, 23-180 ppm; channel catfish; at hatching; hard water) 22 ppm (95% confidence interval, 19-25 ppm; channel

catfish; 4 days post-hatching; hard water)

178 ppm (95% confidence interval, 131-242 ppm; goldfish; at hatching; soft water)

46 ppm (95% confidence interval, 32-66 ppm; goldfish; 4 day post-hatching; soft water)

170 ppm (95% confidence interval, 115-251 ppm; goldfish; at hatching; hard water)

75 ppm (95% confidence interval, 50-112 ppm; goldfish; 4 days post-hatching; hard water)

Method: No specific test guideline was reported; however, a scientifically defensible approach was used to conduct the study.

Test animals were exposed to 10-14 concentrations of boric acid. Depending on the sensitivity of the species, tests were initiated at 50-300 ppm boron, and continued at 2- to 10-fold dilutions until LC_1 and LC_{50} values were determined. In all cases, exposure levels of boric acid were based on actual boron content (boron equivalents), and verified by the chemical analysis of culture water.

Boron treatment was initiated subsequent to fertilization and maintained continuously through 4 days post-hatching, giving exposure periods in days of 28, 9, and 7 for trout, catfish, and goldfish. Minimum sample size per test concentration was 125 for fish embryos. In the analyses of test results, embryos were classified as lethals, teratogenic (anomalous) survivors, or normal survivors. Hatchability was based on all embryos that lived to complete the hatching process. Normal survivors were defined as animals free of the debilitating morphological defects that characterized teratogenic survivors. Test responses were expressed as frequencies in experimental populations/frequencies in controls.

Log probit analysis was used to statistically determine the LC_1 and LC_{50} values. The probit regressions were performed with an IBM computer, and were determined in each instance by using animal responses for the full range of test concentrations. When severely truncated survival curves precluded use of the computer program for LC_{50} determinations, the Litchfield-Wilcoxon graphic method was

used. Analysis of variance and the t-test were used to determine statistical significance of differences in toxicity, water hardness levels, and other test variables.

The synthetic culture medium used for the boron bioassays was prepared from distilled, double deionized water, having a conductivity of 0.25 µmhos or less. Routing monitoring was conducted for background contaminants. The basic stock was prepared to give a water hardness level of 50 or 200 ppm (as $CaCO_3$) and a pH of 7.5-8.0. Different levels of hardness were achieved by dilution of the basic culture water.

Boron bioassays were conducted using a continuous flow system. Fish eggs were cultured through 4 days post-hatching in glass chambers, through which test water was perfused at prescribed flow rates. The test substance was administered to a mixing chamber situated ahead of each culture dish, using graduated flow from a syringe pump. Synthetic water was delivered to the mixing chamber by regulated flow from a peristaltic pump. Flow rates from both syringe and peristaltic pumps were monitored by liquid flow meters. The concentration of test substance delivered to the mixing chamber was regulated by adjusting the mixing ratio from the pumping units and/or by varying the concentration of test substance delivered from the syringe pump. Solutions from the 2 channels were mixed with a magnetic stirring bar, and delivered to the culture chamber under positive pressure. For test concentrations of 10 ppm or more, the boron compound generally was added directly to the culture water in the peristaltic pump reservoir, eliminating the need for the syringe pump channel. In such instances, the test substance remained stable at selected test concentrations for 24 hours or more in polyethylene containers, and important test parameters of culture water were not altered upon standing (e.g., pH, hardness).

The aquatic bioassay cultures were maintained in walk-in environmental rooms. Culture water was given continuous aeration in the peristaltic pump reservoirs. For the bioassays, pH ranged from 7.5-7.9. All cultures were monitored at regular daily intervals for temperature, dissolved oxygen, ammonia, water hardness, and pH. Flow rates from peristaltic and syringe pumps were monitored twice daily.

Boron exposure levels were determined at regular daily intervals by analysis of culture water. Test organisms were examined daily to gauge extent and frequency of development, and to remove dead specimens. Control eggs were also cultured.

Particular attention was given to trout embryos, especially during the "green stage." Harmful exposure to artificial light was precluded and cultures were maintained under semi-sterile conditions to minimize occurrences of soft egg disease and fungus. Prior to each use, culture rooms were disinfected and irradiated with UV for 12 hours, and the rooms were maintained under positive pressure. In bioassays with boron toxicant, a flow rate of 200 mL/hour, giving a turnover time of 1.5 hours, was maintained.

Trout eggs and sperm were collected for test purposes by artificial spawning and milking, using methods of Leitritz, 1972. Fertilization was accomplished by mixing sperm and eggs for 15 minutes immediately prior to the onset of boron exposure. For all other aquatic species, fertilized eggs were collected from natural spawn. Boron treatment was initiated up to 2 hours post-spawning for goldfish and catfish.

Quantitative determinations of boron were accomplished using the curcumin method. This technique proved applicable for a concentration range of 0.1-1.0 mg/L. Concentration and dilution of sample water were used to extend the analytical range. All lots of prepared culture water were monitored for possible boron contamination prior to use for bioassay purposes. To avoid possible contamination from borosilicate glassware, Vycor brand evaporating dishes were used for analytical purposes.

GLP: No

Test Substance: Boric acid, ACS grade

Results: The concentration of ammonia was held below 0.1 ppm in all instances. The temperature was 24.7-25.0, 24.8, and 13.3-13.7° for catfish, goldfish, and trout, respectively. Dissolved oxygen was 7.3-7.6, 7.4-7.5, and 9.2-9.6 ppm for catfish, goldfish, and trout, respectively. The pH was 7.5-7.6, 7.6-7.9, and 7.7-7.9 for catfish, goldfish, and trout, respectively. Water hardness was 51.8, 54.4, and 54.1 ppm $CaCO_3$ at 50 ppm in catfish, goldfish, and trout, respectively. Water hardness was 212.0, 207.5, and 204.0 at 200 ppm $CaCO_3$ at 200 ppm in catfish, goldfish, and trout, respectively.

In no instance was background boron contamination detected in the prepared culture water, including that used for the maintenance of control animals. Actual exposure levels <0.1 ppm were not reported, as analysis of standards prepared at 0.05 ppm or less were not fully reproducible. However, in all such cases, boron levels delivered by syringe pumps to mixing chambers were well above the detection limit, and direct analyses were conducted to confirm syringe pump boron concentrations. In addition, flow rates from syringe pumps (boron) and peristaltic pumps (culture water) were monitored at regular intervals, using both flow meters and direct volumetric measurements, to determine actual boron dilution ratios obtained in the mixing chambers. Dividing actual syringe pump boron concentrations by boron dilution ratios, final boron levels delivered to the bioassay test chambers usually were found to be within 3%, and always within 5%, of selected nominal exposure values. This alternative monitoring procedure was shown for trout bioassays, which included boron exposure values of 0.01 ppm (10 µg/L) and 0.001 ppm (1 µg/L). Though such measurements may be used to confirm boron concentrations delivered to test chambers, they do not provide direct measurements on the stability of exposure levels maintained within the bioassay cultures. While it is unlikely that actual boron exposure values could have exceeded the upper limit of variation (5%) shown for water supplied to the cultures, possible losses of boron from culture water, through tissue accumulation or other means, could have resulted in undetectable reductions in exposure concentrations. However, a high degree of culture stability was obtained for boron concentrations as low as 0.1 ppm, as determined by direct analyses of culture water.

Measure concentrations for tests in hard and soft water for trout, catfish, and goldfish are provided in the tables below.

10 November 2003

Rainbow trout

Nominal boron concentration boron (ppm)	Measured boron concentration (ppm) – soft water (50 ppm $CaCO_3$)	Measured boron concentration (ppm) – hard water (200 ppm $CaCO_3$)
0.001	Not reported	Not reported
0.01	Not reported	Not reported
0.1	0.11	0.10
0.5	Not tested	0.47
1	1.00	0.98
5	4.74	4.85
10	9.26	9.40
25	23.50	23.80
50	45.50	48.30
100	94.00	100.20
200	190.00	186.00

Catfish (*Ictalurus punctatus*)

Nominal boron concentration boron (ppm)	Measured boron concentration (ppm) – soft water (50 ppm $CaCO_3$)	Measured boron concentration (ppm) – hard water (200 ppm $CaCO_3$)
0.01	Not reported	Not reported
0.05	Not reported	Not reported
0.1	0.11	Not tested
0.5	0.49	0.53
0.75	Not tested	0.77
1	1.01	0.96
2.5	Not tested	2.33
5	5.42	4.90
7.5	7.43	7.40
10	10.00	9.43
25	24.90	25.10
50	51.40	48.30
75	Not tested	77.70
100	98.30	Not tested
150	151.00	140.00
200	177.00	Not tested
300	306.41	302.00

Goldfish (*Carassius auratus*)		
Nominal boron concentration boron (ppm)	Measured boron concentration (ppm) – soft water (50 ppm $CaCO_3$)	Measured boron concentration (ppm) – hard water (200 ppm $CaCO_3$)
0.05	Not reported	Not reported
0.1	0.1	0.12
0.5	0.49	0.47
1	0.90	0.90
5	5.20	4.50
7.5	7.00	6.80
10	9.20	8.33
25	22.50	32.00
50	48.70	51.30
100	108.00	96.70
200	188.70	191.00
300	288.00	290.00

Expressed in ppm boron at 4 days post-hatching, LC_1 values for trout, catfish, and goldfish were 0.1, 0.5, and 0.6 for boric acid in soft water and 0.001, 0.2, and 0.2 for boric acid in hard water, respectively.

Rainbow trout: Developmental stages of the trout were exposed for 28 days to boron concentrations ranging from 1 ppb to 200 ppm. Average hatching time was 24 days at 13-14°C. From 1 to 200 ppm boron, percent hatchability of trout eggs generally was inversely proportional to exposure level. At 1 ppm boron, hatchability varied from 94-99%. Treatment with low concentrations of boric acid gave more variable results, with 5-8% embryonic lethality occurring at boron concentrations as low as 0.01 ppm. High concentrations of boron were required to produce substantial levels of embryonic mortality. At 200 ppm, hatching frequencies were reduced to 55% and 65% for boric acid in soft and hard water, respectively. Using 50 ppm boron, egg hatchability varied from 77-82% for boric acid. Boric acid produced the highest frequencies of teratogenic survivors, particularly when used at a water hardness level of 200 ppm $CaCO_3$. In the latter case, 51% of those animals that survived 100 ppm boron were grossly defective, and frequencies dropped to 26, 11, and 5% at 1.0, 0.01, and 0.002 ppm boron, respectively. A high incidence of teratogenesis was observed over a broad range of exposure levels, varying from 200 to 1.0 ppm boron. Results were much more variable and less pronounced when boric acid was administered in soft water (50 ppm $CaCO_3$), with 27, 1,

21, and 5% of survivors bearing anomalies at exposure levels of 200, 50, 1.0, and 0.01 ppm boron.

In the trout bioassay, boron exposure level spanned a dilution range of 200,000 without achieving the full range of test responses. The test responses most characteristic of boron toxicity to embryonic and early juvenile stages of the rainbow trout may be summarized as follows: generally, high frequencies of both embryonic and post-embryonic mortality were recorded only at boron concentrations of 50 ppm or more. Embryonic mortality and teratogenesis were the principal boron-induced responses at 50 ppm or less. Though results with boric acid were variable, trout embryos suffered mortality and teratogenesis at frequencies of 10% or more at boron levels as low as 0.01 ppm, and 4-7% of test animals were affected by 0.001 ppm at 4 days post-hatching. While water hardness did not exert a profound influence on boron bioassays with trout, hard water generally increased embryonic lethality and teratogenesis, and soft water increased boron toxicity to post-hatched alevins. The fact that high concentrations (25-200 ppm) were required to consistently produce substantial impairment of test populations indicates that boron compounds are not highly toxic to trout embryos and alevins. However, compared to trace metals (e.g., cadmium and mercury), boric acid is unusual in that it exerts low-level embryopathic effects on trout over a wide span of exposure levels.

Channel catfish: Average hatching time was 5 days at 25-29°C. Boron exposure levels ranged from 0.01-300 ppm in soft and hard water. Percent egg hatchability and percent normal survival at hatching and 4 days post-hatching gave good inverse correlations with boron concentration. Boron (300 ppm) produced lethality or teratogenesis at hatching in 100% of the experimental populations, and normal survival at 4 days post-hatching was only 0-2% at 200 ppm boron. Using boric acid in soft water, frequencies of normal survival at 4 days post-hatching increased to 56, 75, 95, and 99% at concentrations of 150, 10, 1.0, and 0.5 ppm boron, respectively. Normal survival was 100% at and below 0.1 ppm. Treating with boric acid in hard water, normal survival values at 4 days post-hatching were 9, 65, 86, and 98% at 75, 10, 1.0, and 0.01 ppm boron, respectively. Boric acid produced greater impairment of the test population when administered in hard water. Similar to results with trout, embryonic mortality and teratogenesis increased in

hard water. However, differences were observed with post-hatched stages. Catfish fry were somewhat more sensitive to boron in hard water. As in the trout bioassay, boron concentrations of 50 ppm or more were required to produce high levels of post-hatched mortality. However, catfish fry were somewhat more susceptible than were trout alevins to lower concentrations of boron.

The boron dilution ranges required to give test responses varying from 0-100% were approximately 3000 and 30,000 for boric acid in soft and hard water, respectively. The effective dilution range increased with the order of toxicity. Catfish stages collectively were less sensitive to boron than trout stages by approximate factors of 200 and 5 for boric acid in hard and soft water, respectively. Differential sensitivity was greatest under conditions that proved most toxic to both species (e.g., boric acid in hard water).

Goldfish: Average hatching time was 3 days at 25-27°C. Hatchability and normal survival gave good inverse correlations with boron concentration. Boron at 200 ppm produced complete lethality in all tests. Treating with boric acid in soft water, frequencies of normal survival at 4 days post-hatching were 4, 52, 94, and 98% for boron concentrations of 100, 50, 10, and 1 ppm. With hard water, normal survival averaged 325, 67, 85, and 98% for the same exposure levels.

As for trout, substantial levels of post-hatched mortality were found only at the higher boron concentrations, and post-hatched stages were somewhat more susceptible to boric acid in soft water. Unlike the trout and catfish, appreciable frequencies of teratogenesis occurred only at high exposure levels. Frequencies exceeding 10% were observed only at or above 100 ppm boron.

In all instances, normal post-hatched survival was 92% or more at and below 7.5 ppm boron, and there was a broad near-threshold range extending to 0.05 ppm boron at which low levels of embryonic mortality and/or teratogenesis consistently were observed.

The effective dilution range was approximately 6000 for boric acid. Goldfish were less sensitive than trout stages by approximate factors of 200 and 6 for boric acid in hard and soft water, respectively. As for catfish and trout, the greatest

differential in sensitivity occurred under conditions found most toxic to trout and goldfish.

Reference: Birge, W. J. and J. A. Black (1977). NTIS PB-267085.

Leitritz, E. (1972). Trout and Salmon Culture, State of California, Dept. Fish and Game, Fish. Bull. #107 (cited in Birge and Black, 1977).

Reliability: High because a scientifically defensible or guideline method was used.

Additional References for Acute Toxicity to Fish:

Supporting Data: Isopropyl Alcohol

Data from these additional sources support the study results summarized above. These studies were not chosen for detailed summarization because the data were not substantially additive to the database.

Exxon Corp. (1982). EG&G Report No. BP-82-7-71, TSCA Fiche OTS0510683.

Atlantic Richfield Company (1976). Industrial Bio-Test laboratories, Inc., IBT No. 621-08200 (February 3) (TSCA Fiche OTS0513281).

Exxon Corporation (1982). Bionomics Report #BW-82-7-1226 (July) (TSCA Fiche OTS0510679).

Mattson, V. R. et al. (1976) Ecol. Res. Ser. EPA-600/3-76-097 (AQ-0057190 through AQ-0057194).

Exxon Corporation (1982). EG&G, Bionomics Report #BW-82-7-1229 (July) (TSCA Fiche OTS0510684).

Wolverton, B. C. et al. (1970). Technical Report AFATL-TR-70-68, AD879811.

Brooke, L. T. et al. (eds.) (1984). Acute Toxicities of Organic Chemicals to Fathead Minnows (*Pimephales promelas*), pp. 69-74, Center for Lake Superior Environmental Studies, University of Wisconsin-Superior, Superior, WI.

Supporting Data: Boric Acid

Data from these additional sources support the study results summarized above. These studies were not chosen for detailed summarization because the data were not substantially additive to the database.

Office of Pesticide Programs (1995). Environmental Effects Database (EEDB), Environmental Fate and Effects Division, US EPA, Washington, DC

(AQ-0198457 and AQ-0198458).

Office of Pesticide Programs (1995). Environmental Effects Database (EEDB), Environmental Fate and Effects Division, US EPA, Washington, DC (AQ-0198452 and AQ-0198453).

Procter and Gamble Company (n.d.). Unpublished Report (cited in IUCLID (2000). IUCLID DataSet, "Boric acid, crude natural, containing not more than 85% of H_3BO_3 calculated in the dry weight" (February 18)).

Procter and Gamble Company (1987). Report for the US EPA, Washington, DC (cited in IUCLID (2000). IUCLID DataSet, "Boric acid, crude natural, containing not more than 85% of H_3BO_3 calculated in the dry weight" (February 18)).

4.2 Acute Toxicity to Invertebrates

No data for triisopropylborate exists, since the chemical rapidly hydrolyzes to boric acid and isopropanol. Therefore, for this robust summary, supporting data for the hydrolysis products are presented.

Data for Hydrolysis Product: Isopropanol

Type:	**96-hour LC_{50}**
Species:	*Mysidopsis bahia*, mysid shrimp
Value:	96-hour LC_{50} = 4050 ppm (95% confidence limits, 2530-5030 ppm)
Method:	No specific test guideline was reported; however, a scientifically defensible approach was used to conduct the study.

Mysid shrimp (3-4 days old) were exposed to nominal concentrations of 0, 625, 1250, 2500, 5000, 10,000 or 20,000 ppm in a static 96-hour acute toxicity test. Ten shrimp were tested per dish and treatments were duplicated, resulting in 20 shrimp per treatment. There was no aeration. Dissolved oxygen (DO) concentrations were $\geq$87% of saturation at initiation of the test. Shrimp were fed live brine shrimp nauplii on days 0 and 2 of the test.

Test concentrations were prepared by adding appropriate volumes of the test substance to the seawater in the test containers. The test containers were then covered with aluminum foil and stirred for 4 hours. The shrimp were added to the test containers when the temperature decreased to 23°C. A concurrent seawater control was conducted.

	Water samples for chemical analysis were taken from each test container at the initiation and termination of the test. If all shrimp died in a test container prior to termination of the test, the water sample was taken at that time. All water samples were refrigerated until shipment for analysis. Based on the results of the test, the LC_{50} values were calculated using the moving average angle method.
GLP:	Unknown
Test Substance:	Isopropanol, purity not reported
Results:	After 3 hours of exposure, mortality was 0% in all test concentrations. After 96 hours of exposure, mortality ranged from 5% in test concentrations $\leq$2500 ppm to 100% in the 10,000 and 20,000 ppm test concentrations. There was 5% control mortality. The 24- 48-, and 72-hour LC_{50}s were 5910, 4960, and 4170 ppm, respectively. The test salinity and temperature were 20o/oo and 21-23°C. The dissolved oxygen concentrations and pH remained within acceptable ranges throughout the test. After 96 hours of exposure, dissolved oxygen concentrations were >87% of saturation in all test concentrations and the control. Initial pH was 8.3-8.5 in the control and all test concentrations. The pH after 96 hours of exposure was 7.9-8.0 in the control and all test concentrations with surviving shrimp.
Reference:	Exxon Corp. (1982). EG&G Bionomics Report No. BP-82-7-69, TSCA Fiche OTS0510681.
Reliability:	Medium because a suboptimal study design (nominal test concentrations) was used.
Type:	**24-hour EC_{50}**
Species:	*Daphnia magna*
Value:	159,000 µmol/L (29,906 mg/L)
Method:	Test procedures were based on OECD Guideline 202. The 24-hour EC_{50} was calculated by the Trimmed Spearman-Karber method.
GLP:	Unknown
Test Substance:	Isopropanol, purity >97%
Results:	No additional data were reported.
Reference:	Calleja, M. C. et al. (1994). Arch. Environ. Contam. Toxicol., 26:69-78.
Reliability:	Medium because a suboptimal study design (nominal test concentrations) was used.

Type:	**16-day log NOEC (growth)**
Species:	*Daphnia magna*
Value:	3.37 µmol/L (0.63 mg/L)
Method:	No specific test guideline was reported; however, a scientifically defensible approach was used to conduct the study. *Daphnia* (<1 day old) were exposed to the test substance for 16 days (3-4 broods) in 2 duplicate experiments with 15 organisms each. The test volume was 1 L/group, and the test medium was Dutch standard water at a hardness of ca. 1 mmol/L. The static test had a ratio between the concentrations of the toxicant tested of 1.8, and the renewing rate was 3 times/week. *Daphnia* were fed *Chlorella* sp. during the test. At the start of the experiments and after 16 days, the lengths of 30 daphnids were measured. The NOEC on growth was defined as the highest concentration that did not result in a significant reduction in growth (tested with Student's t-test). During the test, the actual concentrations were determined once for the lowest and highest concentration just before and after renewal of the test solutions in the semi-static experiments.
GLP:	Unknown
Test Substance:	Isopropanol, purity not reported
Results:	The chemical analyses showed that between 80 and 110% actually was present in the test solutions. The percentage was a mean value of analyses from samples out of the duplicate experiments that were taken just before and after the test solutions were renewed. The NOEC value was not corrected for the measured concentration, since this had only a slight influence on the calculated QSARs.
Reference:	Hermens, J. et al. (1985). Aquatic Toxciol., 6:209-217. Hermens, J. et al. (1984). Aquatic Toxicol., 5:143-154.
Reliability:	High because a scientifically defensible or guideline study was used.

Type:	**16-day log EC_{50} (reproduction)**
Species:	*Daphnia magna*
Value:	4.73 µmol/L (0.89 mg/L)
Method:	No specific test guideline was reported; however, a scientifically defensible approach was used to conduct the study.

	Daphnia (<1 day old) were exposed to the test substance until the control daphnids had produced 4 broods). The test volume was 1 L/group, and the test medium was Dutch standard water at a hardness of ca. 1 mmol/L. The static test had a ratio between the concentrations of the toxicant tested of 1.8, and the renewing rate was 3 times/week. *Daphnia* were fed *Chlorella* sp. during the test. Fifteen daphnids were exposed to each test concentration in duplicate experiments. In the semistatic tests on single compounds, the actual concentrations were determined once for the lowest and highest concentration (in duplicate) just after and before renewal of the test solutions. The test substance was analyzed using gas-liquid chromatography. The NOEC on reproduction was defined as the highest concentration that did not result in a reduction of reproduction compared with the control. The EC_{50} values were calculated by the method of Litchfield and Wilcoxon. The NOEC on growth was defined as the highest concentration that did not result in a significant reduction in growth compared with the control.
GLP:	Unknown
Test Substance:	Isopropanol, purity not reported
Results:	The log NOEC for reproduction and growth were 4.37 and 4.11 µmol/L, respectively. Determined concentrations were 80-104% of the calculated amount. The percentages were mean values of at least 4 determinations. The average decrease in concentration during the tests was 9%, with a maximum of 26%. The NOEC and EC_{50} values were not corrected for the measured losses because this would have only a slight influence on the QSARs.
Reference:	De Wolf, W. et al. (1988). Aquatic Toxicol., 12:39-49. Hermens, J. et al. (1984). Aquatic Toxicol., 5:143-154.
Reliability:	High because a scientifically defensible or guideline method was used.

Data for Hydrolysis Product: Boric Acid

Type:	**48-hour LC_{50}** **21-day LC_{50}**
Species:	*Daphnia magna*

10 November 2003

Value: 48-hour LC_{50} = 133 mg/L, as boron (95% confidence interval, 115-153 mg/L)

21-day LC_{50} = 52.2 mg/L, as boron (95% confidence interval, 42.6-66.7 mg/L)

Method: Acute toxicity testing procedures were based on the guidelines of the ASTM Subcommittee on Safety to Aquatic Organisms, Standard E 729-80.

Lake Huron water was used to culture daphnids and as test dilution water. The water was carbon-filtered, UV-irradiated, and adjusted to a hardness of approximately 170 mg/L as $CaCO_3$. The water was then autoclaved at 120°C and 124.1 kPa for 35 minutes.

Acute Toxicity Test: The static acute test was conducted in 250 mL beakers to which 200 mL of the appropriate amount of test substance and water was added. The test consisted of exposing 3 replicate groups of 10 neonates to each of 6 nominal concentrations (54, 91, 151, 252, 420, and 700 mg/L as boron) of the test substance and control. In addition, an extra beaker was set at the high, medium, low, and control concentrations to avoid the risk of contamination while taking dissolved oxygen, pH, and temperature measurements. The test beakers were placed in a temperature-controlled environmental chamber set at 20±1°C and a photoperiod of 16 hours light:8 hours dark. The duration of the test was 48 hours. Mortality, as well as dissolved oxygen, pH, and temperature were recorded after 24 and 48 hours of exposure. Daphnids were not fed, nor were the solutions aerated during the test.

Chronic Toxicity Test: A static renewal procedure, with batchwise replacement of test and control solutions at regular intervals (Monday, Wednesday, and Friday) was used in the chronic test. The test vessels were 600 mL glass beakers. Each beaker contained a mesh stainless steel platform and 5 glass tubes with nylon mesh bottoms. The tubes were placed on the platform to allow water circulation. Each test beaker contained 500 mL of the appropriate amounts of test material, food, and water. During the test the solutions were gently aerated to achieve 90 to 105% saturation. There were 4 replicates for each test concentration and the control, resulting in 5 daphnids per replicate or a total of 20 organisms per concentration. The daphnids were fed *S. capricornutum* during the test. The test

beakers were placed in a temperature-controlled environmental chamber set at 20±1°C and a photoperiod of 16 hours light:8 hours dark. The duration of the study was 21 days. The chronic study began by placing one neonate in each tube, where the daphnid remained for the entire study. Each Monday, Wednesday, and Friday the young produced by each adult were counted and discarded, and adult survival was recorded. In addition, on the same days, the dissolved oxygen, pH, and temperature in each test concentration and the control were measured and recorded. After enumeration of the young, the adults were transferred to clean beakers containing fresh test and control solutions, in addition to a new supply of food. The boric acid test concentrations used for the chronic test were 7, 14, 28, 56, and 105 mg/L as boron. The test concentrations were verified using the carmine method. On each Monday, Wednesday, and Friday analyses were performed on all replicates from 1 particular test concentration and on 1 replicate from each remaining test concentration and control. The boron concentrations were also analyzed in renewed solutions (time zero) and the same test solution prior to the next renewal to determine the stability of the test substance.

The LC_{50} value for the acute toxicity test was based on nominal concentrations, but the LC_{50} value for the chronic toxicity test was based on measured concentrations. Finney's method of probit analysis or the moving average method was used to calculate the LC_{50} values. Data from the chronic portion of the study were analyzed using a two-tailed Dunnett's t test. Mean comparisons between test and control concentrations were performed on the mean number of broods per daphnid, mean total young per daphnid, mean brood size per daphnid, and mean length, in effort to estimate the maximum acceptable toxicant concentration (MATC).

GLP: Unknown

Test Substance: Boric acid, purity not reported

Results: During the study, pH was 8.1±0.1, conductivity was 290±31 µmhos/cm, hardness was 148±7 mg/L as $CaCO_3$, and alkalinity was 58±5 mg/L as $CaCO_3$. The boron concentration in the water was 0.4±0.1 mg/L. Lake Huron contains about 12 µg/L boron.

Acute Toxicity Test: The no-kill level was <54 mg/L and the 100% kill concentration was 420 mg/L. There was 7% control mortality during the 48-hour test. Dissolved oxygen

concentrations throughout the test were greater than 60% saturation and the pH ranged from 6.7 to 8.1. Temperatures ranged from 20.1 to 20.7°C.

Chronic Toxicity Test: The mean boron concentration was 0, 6.4, 13.6, 29.4, and 59.3 at 0, 7, 14, 28, and 56 mg/L, respectively. The means of the analyzed concentrations ranged from 91.4 to 106% of the nominal concentrations. The boron concentrations in the renewed test solutions (time zero) and for the same test solutions prior to the next renewal were not significantly different, thus attesting to the stability of the test substance over the renewal period. Throughout the chronic test the dissolved oxygen and temperature ranged from 7.3-8.0 mg/L and 19.5-20.5°C, respectively. Mortality during the 21-day test was 0, 0, 10, 5, and 40% at 0, 7, 14, 28, and 56 mg/L, respectively. No data were available for daphnids exposed to the highest concentration of boric acid (105 mg/L as boron) because none survived to reproductive age. Time to first reproduction was not affected by the test concentrations. The mean number of broods per daphnid, mean total young per daphnid, mean brood size per daphnid, and mean size all differed significantly from control at 13.6 mg/L. It appeared that the most biologically, as well as statistically, important endpoints for this study were those associated with reproduction and growth. Therefore, determination of the MATC was based on the endpoints of mean total young per replicate, mean brood size, and mean size. The MATC of boric acid was estimated to lie between 6.4 and 13.6 mg/L as boron.

Reference: Gersich, G. M. (1984). Environ. Toxciol. Chem., 3:89-94.

Reliability: High because a scientifically defensible or guideline method was used.

Type: **8-day EC_{50}**

Species: *Ceriodaphnia dubia*

Value: >100 mg/L

Method: The definitive toxicity test was performed according to U.S. Environmental Protection Agency Method 1002.0, "Short Term Method for Estimating the Chronic Toxicity of Effluents and Receiving Waters to Freshwater Organisms. *Cerodaphnia* Survival and Reproduction Test," EPA/600/4-85/014.

The test was performed under static, renewal conditions at nominal concentrations of 0, 6.25, 12.5, 25, 50, and 100 mg/L. The test was conducted at a target temperature of

25±1°C. Stock solutions were prepared every day during the test. The test media was renewed in each test vessel daily. Dilution water was bottled drinking water adjusted to a hardness of approximately 100 mg/L as $CaCO_3$. Water was stored in a glass tank where it was temperature adjusted and aerated.

Ten neonates, less than 24 hours old at test initiation, were indiscriminately distributed among 10 replicates of each concentration (1 per vessel). Adults were transferred to fresh dilution water every 1-8 days. The culture was maintained in a 4-L glass jar that was filled to approximately 50-90% capacity. The culture was supplied with a yeast/trout chow mix and freshwater alga daily before the start of the test.

Test vessels were arranged in an incubator. A 16-hour light and 8-hour dark photoperiod was automatically maintained with cool-white fluorescent lights. Aeration was not used.

The number of surviving adults and the occurrence of sublethal effects (immobilization, loss of equilibrium, erratic swimming, loss of reflex, excitability, discoloration, or change in behavior) were determined visually and recorded at test start and every 24 hours. The young produced by each adult were counted and removed daily after the onset of reproduction.

Dissolved oxygen, pH, conductivity, and temperature were measured and recorded daily before and after media renewal in a composite of media from each test chamber. The hardness and alkalinity of the dilution water control and the highest tested concentration were measured and recorded daily.

All calculations were performed using nominal concentrations of test substance. Survival of first generation adults was not statistically analyzed and the EC_{50} could not be determined because 100% survival occurred at all tested concentrations. The number of young per adult was statistically analyzed. A Chi-square test was used to determine that data were normally distributed, and Bartlett's test was used to determine that variances were homogeneous. Because the assumption of homogeneity of variances was met, a parametric one-way analysis of variance followed by Dunnett's test was used to compare

	treatment with control means. Data were not transformed prior to statistical analyses.
GLP:	Yes
Test Substance:	Boric acid, purity >99%
Results:	A screening test was performed for 48-hours to nominal concentrations of 0, 0.10, 1, 10, 100, and 1000 mg/L. At the end of the test there was 0% survival at 1000 mg/L, 10% survival at 100 mg/L, and 100% survival at all lower concentrations. Insoluble material was not observed in any test vessel during the test. Control survival was 100% at test end and first-generation adults produced an average of 17.4 young during the 3-brood study. Water quality parameters were within acceptable limits throughout the study. Dissolved oxygen concentrations were always above 7.8 mg/L. During the test, the mean temperature was 24.4°C, the mean conductivity was 350 µmhos/cm, and the pH ranged from 7.1 to 8.3. Hardness ranged from 88 to 100 mg/L in control vessels and from 88 to 108 mg/L in vessels containing 100 mg/L of the test substance. Alkalinity ranged from 16 to 21 mg/L in control vessels and from 16 to 23 mg/L in vessels containing 100 mg/L of the test substance. The percentage of surviving adults was not significantly reduced at any tested concentration, and sublethal effects were not observed at any concentration. The mean number of young per surviving adult was significantly lower than the control at the 3 highest tested concentrations (25, 50, and 100 mg/L). The most sensitive biological endpoint was production of young. The NOEC, LOEC, and MATC were12.5, 25, and 17.7 mg/L, respectively. The 8-day EC_{50} was >100 mg/L. Because no adults were affected, the EC_{50} response criteria was death.
Reference:	DuPont Co. (1993). Unpublished Data, Haskell Laboratory Report HLO-418-93, "Chronic Toxicity to *Ceriodaphnia dubia*" (June 2).
Reliability:	Medium because a suboptimal study design (nominal test concentrations) was used.
Type:	**14-day NOEL**

10 November 2003

Species:	*Daphnia mgana*
Value:	Approximately 14 mg/L
Method:	No specific test guideline was reported; however, a scientifically defensible approach was used to conduct the study.

All daphnids were cultured and tested in Lake Huron water, which was adjusted to a hardness of approximately 170 mg/L as $CaCO_3$, prior to autoclaving. The water was autoclaved at 121°C and 124 kPa for 35 minutes. All daphnids were fed a diet of *Selenastrum capricornutum* Printz.

The chronic tests were conducted in a manner similar to that reported by Gerisch, 1984 and Gerish et al., 1984. The studies were designed to use a static renewal procedure with batchwise replacement of the test and control solutions on a Monday, Wednesday, and Friday basis. Two chronic daphnid studies were conducted. The test vessels were 600 mL glass beakers, each containing 5 glass tubes with wire mesh bottoms. The tubes were supported off the bottom of the beaker with a mesh stainless steel platform. During the studies, each beaker contained the appropriate amount of food, dilution water, and test material made up to a 500 mL volume. The test organisms were fed *Selenastrum capricornutum*. The beakers were held in a temperature controlled environmental chamber set at 24±2°C and a photoperiod of 16 hours daylight/8 hours darkness.

The chronic tests began by placing 1 neonate daphnid in each uniquely labeled tube. The daphnids remained in their respectively labeled tubes for the duration of the study. Each test and control concentration had 4 replicates, resulting in 20 daphnids being exposed to each concentration. The duration of each test was 14 days. The critical endpoints were associated with reproduction, growth, and survival. The reproductive endpoint was obtained by hand counting the neonates on a Monday, Wednesday, and Friday basis. Growth was calculated by determining the dry weight of the surviving adult organisms at the tests end. Survival data were collected every Monday, Wednesday, and Friday. Additionally, pH, dissolved oxygen, and temperature were measured and recorded on each renewal day.

Samples from the boric acid chronic static renewal tests were analyzed, using an appropriate high performance liquid

chromatography method. On each Monday, Wednesday, and Friday during all studies, analyses were performed on a replicate from each test concentration and the control.

Data derived from the studies were analyzed by a one-tailed Dunnett's test. The Dunnett's procedure used simultaneously tested for heterogeneity of variances using Bartlett's test. If the variances were heterogeneous, the Wilcoxon signed rank test was used to compare the means. Mean comparisons between test and control concentrations were performed on percent survival, meant total young/adult, mean brood size/adult, and mean dry weight/adult to estimate the MATC.

GLP: Unknown

Test Substance: Boric acid, purity 99.5%

Results: During both boric acid studies, all analyzed concentrations were within a range of 98.5 and 111.4% of nominal.

Interpretation of the chronic data for boric acid indicates that the MATC is between 13.8 and 28.1 mg/L for Test I and between 14.3 and 28.9 mg/L for Test II. Expressing the MATC as a geometric mean of these concentrations for Tests I and II resulted in MATC's of 19.7 and 20.3 mg/L, respectively. The NOEL for both studies was approximately 14 mg/L. The estimation of the MATC's was based on data associated with survival, reproduction, and growth. These endpoints all significantly differed from the controls at the 28 mg/L concentration. No other toxic effects were observed during the studies. During both studies, >48% of the healthy organisms had first broods on day 6.

During the two 14-day studies with boric acid, no mortality was observed in the controls. The dissolved oxygen measurements were >90% saturation during both tests (range 8.3 to 8.8 mg/L). The pH and temperature measurements ranged from 7.3 to 8.2 and 23.0 to 25.2°C, respectively.

Reference: Gersich, F. M. and D. P. Milazzo (1990). Arch. Environ. Contam. Toxicol., 19:72-76.

Gerish, F. M. (1984). Environ. Toxicol. Chem., 3:89-94 (cited in Gersich, F. M. and D. P. Milazzo (1990). Arch. Environ. Contam. Toxicol., 19:72-76).

Gerisch, F. M. et al. (1984). Bull. Environ. Contam. Toxicol., 32:497-502 (cited in Gersich, F. M. and D. P. Milazzo (1990). Arch. Environ. Contam. Toxicol.,

19:72-76).

Reliability: High because a scientifically defensible or guideline method was used.

Additional References for Acute Toxicity to Invertebrates:

Supporting Data: Isopropanol Alcohol

Data from these additional sources support the study results summarized above. These studies were not chosen for detailed summarization because the data were not substantially additive to the database.

Blackman, R. A. A. (1975). Mar. Pollut. Bull., 1:116-118.

Lilius, H. et al. (1994). Aquatic Toxicol., 30:47-60.

Lilius, H. et al. (1994). Environ. Toxicol. Chem., 14(12):2085-1088.

Bringmann, G. and R. Kühn (1982). Z. Wasser Abwasser Forsch., 15(1):1-6.

Bringmann, G. and R. Kühn (1977). Z. Wasser Abwasser Forsch., 10(5):161-166.

McCauley, D. J. (n.d.). Data, Wisconsin Department of Natural Resources, Madison, WI (cited in Vaishnav, D. D. and E. T. Korthals (1990). Arch. Environ. Contam. Toxicol., 19:624-628).

Supporting Data: Boric Acid

Data from these additional sources support the study results summarized above. These studies were not chosen for detailed summarization because the data were not substantially additive to the database.

Dow Chemical Co. (2000). TSCA Fiche OTS0518450.

Office of Pesticide Programs (1995). Environmental Effects Database (EEDB), Environmental Fate and Effects Division, US EPA, Washington, DC (AQ-0198454 through AQ-0198456 and AQ-0200581).

4.3 Acute Toxicity to Aquatic Plants

No data for triisopropylborate exists, since the chemical rapidly hydrolyzes to boric acid and isopropanol. Therefore, for this robust summary, supporting data for the hydrolysis products are presented.

Data for Hydrolysis Product: Isopropanol

Type:	**5-Day Algistatic Concentration (AC)**
Species:	*Selenastrum capricornutum*, green algae
Value:	54,294 ppm (95% confidence limits, 41,565-70,922 ppm)
Method:	Test procedures were based on "A Method for Measuring Algal Toxicity and Its Application to the Safety Assessment of New Chemicals (Payne and Hall, 1979), and the U.S. Environmental Protection Agency (1978). A primary stock solution was prepared by adding a weighed amount of test material to algal growth medium. The test concentrations were then prepared by adding appropriate volumes of the primary stock solution to the test containers. Test conditions included a temperature of 24±1°C and light intensity of approximately 4000 lux. Algae were tested at nominal concentrations of 3125, 6250, 12,500, 25,000, and 50,000 mg/L for five days. The effect criterion was change in cell numbers.
GLP:	Unknown
Test Substance:	Isopropanol, purity 100% active ingredient
Results:	After 5 days of exposure, the percentage change of cell numbers in exposed cultures as compared to the control was from +121% at 3125 ppm to –99% in cultures exposed to 50,000 ppm. Measurements of *in vivo* chlorophyll *a* showed a growth-concentration response similar to the observed effect based on cell numbers. After 5 days of exposure, the percentage change of relative fluorescence units was from +17 in cultures exposed to 3125 ppm to –100% in cultures exposed to 50,000 ppm. Based on cell numbers, the growth of cultures previously exposed to 50,000 ppm was approximately one-half (-47%) that of the controls during a 9-day recovery period in test material-free medium, indicating at least a partial algicidal effect. Measurements of *in vivo* chlorophyll *a* showed a growth response similar to the observed effect based on cell numbers during the recovery phase. After 9 days of recovery, the percentage decrease was 50% in the previous 50,000 ppm exposure concentration when compared to the growth medium control.
Reference:	Exxon Corp. (1983). EG&G Report No. BP-83-2-9, TSCA Fiche OTS0510685.
Reliability:	Medium because a suboptimal study design (nominal test concentrations) was used.

Data for Hydrolysis Product: Boric Acid

Type:	**Boron Tolerance and Accumulation**
Species:	*Lemna minor* L., duckweed
Value:	7-day NOEC > 20 µg/mL

Method:	No specific test guideline was reported; however, a scientifically defensible approach was used to conduct the study. Cultures of duckweed were maintained axenically in F-medium, which was adjusted to pH 5. The plants were grown in continuous 100 µE/m^2/s mixed cool-white fluorescent and incandescent light in the 400 to 700 nm region at 25±2°C, and transferred as three-frond clones of approximately the same age to sterile F-medium incorporating boric acid (10 to 20 µg/mL). Despite the high pK's for this weak acid, it was in all cases soluble. Replicate cultures (12-15) were incubated for a typical 7-day experiment. Daily frond number counts for each replicate were made and the change in frond number was calculated. The frond doubling time was calculated using a procedure to ensure growth rate only during the steady-state phase was measured – lag period effects were avoided by excluding data of day 1 and stationary phase effects were not apparent during the treatment period. Plants were harvested and washed, towel-dried, weighed, and hand homogenized. The homogenates were centrifuged and aliquots of the supernatant assayed for boron. The insoluble pellet was digested, diluted, and centrifuged, and protein determined in the supernatant by the Folin method. Boron was determined colorimetrically in the soluble extract after reaction with Azomethine-H. Azomethine-H asays were run in duplicate on triplicate plant samples of each treatment comprising an experiment, and each experiment repeated at least twice.
GLP:	Unknown
Test Substance:	Boric acid, purity not reported
Results:	*Lemna minor* tolerated between 10 and 20 µg/mL elemental boron in the growth medium at pH 5.0 without being inhibited. Plants grown for 6 days in 50 or 100 µg/mL boron in the medium partially recovered control rates of growth during 5 days incubation in control medium; 200 µg/mL boron was toxic beginning at about 3 days of exposure.
Reference:	Frick, H. (1985). J. Plant Nutr., 8(12):1123-1129.
Reliability:	High because a scientifically defensible or guideline method was used.

Additional References for Acute Toxicity to Aquatic Plants:

Supporting Data: Isopropyl Alcohol

Data from this additional sources support the study results summarized above. This study was not chosen for detailed summarization because the data were not substantially additive to the database.

Adams, V. D. et al. (1975). NTIS PB250730.

Supporting Data: Boric Acid

Data from these additional sources support the study results summarized above. These studies were not chosen for detailed summarization because the data were not substantially additive to the database.

Wong, P. K. and C. K. Wong (1990). Bull. Environ. Contam. Toxicol., 45:752-759.

Anita, N. J. and J. Y. Cheng (1975). J. Fish. Res. Board. Can., 32:2487-2494 (cited in IUCLID (2000). IUCLID DataSet, "Boric acid, crude natural, containing not more than 85% of H_3BO_3 calculated in the dry weight" (February 18)).

Nobel, W. (1981). Angewandte Botanik, 55:501-514 (AQ-0066917 through AQ-0066924).

5.0 Mammalian Toxicity

5.1 Acute Toxicity

Type:	**Oral LD_{50}**
Species/Strain:	Male rats/ChR-CD
Value:	8126 mg/kg (95% confidence limits, 7828-8549 mg/kg)
Method:	No specific test guideline was reported; however, a scientifically defensible approach was used to conduct the study. The test substance, as received, was administered by intragastric intubation in single doses of 7000, 7500, 8000, and 9000 mg/kg, to 4 groups of young adult male rats, 10 animals per dose level. Survivors were sacrificed without pathologic examination 14 days later. The LD_{50} was calculated from mortality data using the method of D. J. Finney.
GLP:	No
Test Substance:	Triisopropylborate, pure
Results:	Mortality was 0/10, 1/10, 5/10, and 9/10 at 7000, 7500,

8000, and 9000 mg/kg, respectively. All mortality occurred 1-3 days after dosing. Weight loss occurred 1-3 days after dosing at all levels tested. Test days that clinical signs were observed are included in the table below.

Dose (mg/kg):	7000	7500	8000	9000
Clinical Sign:				
Alopecia	-[a]	-	2	-
Belly to cage posture	0	0	0-2	0
Chromodacryorrhea	1-3	1-3	1,3-5	3-6,8
Diarrhea	2-3	2-3	2-5	-
Exophthalmos	3	-	-	-
Labored breathing	-	1	-	1-2
Lacrimation	-	1	-	0-2
Lethargic	-	0	-	3
Moribund	-	1	-	1-2
Piloerection	2-5,12	2-3,8	4-5,10	14
Prostration	0	0,2	0	-
Rapid breathing	1	0	1	0,4
Stained nose/face/mouth	-	-	1-3	1-3
Stained perineal	-	-	3	-
Unkempt fur	2	8	-	5-6
Weakness	1	-	1	-
[a] Clinical sign was not observed in any rat at this dose level.				

Reference: DuPont Co. (1978). Unpublished Data, Haskell Laboratory Report No. 491-78, "Oral LD_{50} Test" (August 25).

Reliability: High because a scientifically defensible or guideline method was used.

Type: **Oral LD_{50}**

Species/Strain: Mice/Swiss

Value: 2500 mg/kg (2.5 mL/kg)

Method: No specific test guideline was reported.

The acute oral toxicity of the test substance was determined by forced feeding of the test substance to adult Swiss mice. From the dose that killed 50% of the animals during a 10-day observation period and the weight of the animals, the LD_{50} dose was calculated. No information regarding vehicle, gender, number of mice per dose level, or dose levels tested was reported.

GLP: No

Test Substance:	Triisopropylborate, purity not reported
Results:	No additional data were reported.
Reference:	Adams, R. M. (ed.) (1964). Boron, Metallo-boron Compounds, and Boranes, pp. 693-737, Interscience Publishers, New York (also cited in RTECS/ED5950000).
Reliability:	Not assignable because limited study information was available.

Additional Reference for Acute Oral Toxicity:

Data from this additional source support the rat LD_{50} study results summarized above. This study was not chosen for detailed summarization because the data were not substantially additive to the database.

DuPont Co. (1975). Unpublished Data, Haskell Laboratory Report No. 512-75, "Acute Oral Test" (August 15).

Type:	**Inhalation Toxicity:** No Data.

Type:	**Dermal Toxicity:** No Data.

Type:	**Dermal Irritation**
Species/Strain:	Male guinea pigs/Albino
Method:	No specific test guideline was reported; however, a scientifically defensible approach was used to conduct the study. The test for primary irritation was conducted by applying and lightly rubbing in one drop (approximately 0.05 mL) each of the test substance as received and as a 25% solution (vol/vol) in distilled water on the shaved, intact shoulder skin of 10 male albino guinea pigs (initial average weight 441 g).
GLP:	No
Test Substance:	Triisopropylborate, purity not reported
Results:	The test substance did not produce primary irritation in any guinea pig tested at 100% or 25%. One guinea pig in the 100% group died due to non-test substance-related causes.
Reference:	DuPont Co. (1975). Unpublished Data, Haskell Laboratory Report No. 501-75, "Primary Skin Irritation and Sensitization Tests on Guinea Pigs" (August 28).
Reliability:	High because a scientifically defensible or guideline method was used.

Additional References for Dermal Irritation: None Found.

Type:	**Dermal Sensitization**
Species/Strain:	Male guinea pigs/Albino
Method:	No specific test guideline was reported; however, a scientifically defensible approach was used to conduct the study. The test for primary irritation was conducted by applying and lightly rubbing in one drop (approximately 0.05 mL) each of the test substance as received and as a 25% solution (vol/vol) in distilled water on the shaved, intact shoulder skin of 10 male albino guinea pigs (initial average weight 441 g). To test for the sensitization potential, a series of 4 sacral intradermal injections was given, 1 each week over a 3-week period, which consisted of 0.1 mL of a 1% solution (vol/vol) of the test substance in physiological saline. Following a 2-week rest period, the test animals were challenged for sensitization by applying, and lightly rubbing in, 1 drop (approximately 0.05 mL) each of the material as received and as a 25% solution (vol/vol) in distilled water on the shaved intact shoulder skin. A group of 10 previously unexposed guinea pigs (average weight 664 g) received similar applications at the time of challenge to provide a direct comparison of the challenge reactions on skin of similar age.
GLP:	No
Test Substance:	Triisopropylborate, purity not reported
Results:	The test substance did not produce primary irritation or sensitization in any guinea pig tested at 100% or 25%. One guinea pig in the 100% group died due to non-test substance-related causes.
Reference:	DuPont Co. (1975). Unpublished Data, Haskell Laboratory Report No. 501-75, "Primary Skin Irritation and Sensitization Tests on Guinea Pigs" (August 28).
Reliability:	High because a scientifically defensible or guideline method was used.

Additional References for Dermal Sensitization: None Found.

Type:	**Eye Irritation**
Species/Strain:	Rabbits/Albino
Method:	No specific test guideline was reported; however, a scientifically defensible approach was used to conduct the study. One-tenth milliliter of the undiluted test substance was placed into the right conjunctival sac of each of 2 albino

	rabbits. After 20 seconds, 1 treated eye was washed with tap water for 1 minute. The treated eye of the other rabbit was not washed. Observations of the cornea, iris, and conjunctiva were made with a hand-slit lamp at 1 and 4 hours, and at 1, 2, and 3 days. Fluorescein stain and a biomicroscope were used at examinations after the day of treatment.
GLP:	No
Test Substance:	Triisopropylborate, purity not reported
Results:	The test substance produced no corneal, iritic, or conjunctival effects in the rabbit eye. Both treated eyes were normal at 1 hour.
Reference:	DuPont Co. (1975). Unpublished Data, Haskell Laboratory Report No. 500-75, "Eye Irritation Test in Rabbits" (August 28).
Reliability:	High because a scientifically defensible or guideline method was used.

Type:	**Eye Irritation**
Species/Strain:	Rabbits/Strain not reported
Method:	No specific test guideline was reported. A dose consisting of 100 mg of the undiluted liquid was placed in the conjunctival sac of the eye. Eyelids were held together for at least 1 minute. Observations were made for at least 1 week following dosing.
GLP:	No
Test Substance:	Triisopropylborate, purity not reported
Results:	The test substance produced mild eye irritation in rabbits.
Reference:	Adams, R. M. (ed.) (1964). Boron, Metallo-boron Compounds, and Boranes, pp. 693-737, Interscience Publishers, New York (also cited in RTECS/ED5950000).
Reliability:	Not assignable because limited study information was available.

Additional References for Eye Irritation: None Found.

5.2 Repeated Dose Toxicity: No Data.

5.3 Developmental Toxicity: No Data.

5.4 Reproductive Toxicity: No Data.

5.5 Genetic Toxicity

Type: ***In vitro*** **Bacterial Reverse Mutation Assay**

Tester Strain:	*Salmonella typhimurium* TA98, TA100, TA1535, TA1537, and TA1538
Exogenous Metabolic Activation:	With and without rat liver homogenate activation system (S9)
Exposure Concentrations:	0, 1000, 3000, 5000, 7000, and 10,000 µg/plate
Method:	No specific test guideline was reported; however, a scientifically defensible approach was used to conduct the study.

The assay was performed as 2 independent trials in the presence and the absence of a rat liver homogenate activation system (S9). In the absence of an activation system, a solution of the test substance and approximately 10^8 bacteria in 0.1 mL were added to top agar. These components were mixed and poured on the surface of a plate containing Davis minimal agar. To treat in the presence of an activation system, S9 mix was added to the bacteria-test sample-top agar mixture. S9 was the 9000 x g supernatant of liver homogenate from rats given Aroclor 1254 5 days before sacrifice. The S9 mix contained S9, $MgCl_2$, KCl, glucose-6-phosphate, NADP, and sodium phosphate (pH 7.4). The S9 mix was added to the bacteria, test sample, and top agar. These components were mixed and immediately poured over the minimal agar plate. The revertant colonies were counted after the plates were incubated at 37°C for 48 hours. Positive controls included 2-aminoanthracene, N-methyl-N'-nitro-N-nitrosoguanidine, 9-aminoacridine, and 2-nitrofluorene. The solvent control was acetone.

The cytotoxicity of the test substance in the presence and absence of an activation system, as measured in strain TA1535, was the basis for selecting concentrations to be used in the mutagenesis experiment. The protocol used to determine the cytotoxicity was identical to the mutagenesis protocol, except that 10^3 rather than 10^8 bacteria were used per plate and a nonlimiting concentration of histidine was present. Concentrations of test sample that were nontoxic and, if possible, slightly toxic were selected for the mutagenesis assay.

Data from replicate plates within a single experiment were averaged. The average of those values from different experiments was determined. The highest average number of revertants that was obtained was expressed as a multiple

	of the control value for the sensitive strain(s). When a test sample was active, the average numbers of revertants observed before activity plateaus or decreases at the various concentrations tested were submitted to linear regression analysis. The slope of the line thus obtained was used to determine the number of revertants/nmole or µg of test sample. The test material was classified as nonmutagenic if the reversion frequency was less than 2 times the spontaneous frequency, if less than 0.02 revertants/nmole were observed.
GLP:	No
Test Substance:	Triisopropylborate, pure
Results:	Negative
Remarks:	The initial cytotoxicity experiment with strain TA1535 failed to demonstrate a toxic effect for the test substance at the concentrations tested. Higher concentrations were not included in the mutagenesis experiments. The test substance was not mutagenic in the presence or absence of an activation system.
Reference:	DuPont Co. (1978). Unpublished Data, Haskell Laboratory Report No. 304-78, "Mutagenic Activity in the *Salmonella*/Microsome Assay" (June 9).
Reliability:	High because a scientifically defensible or guideline method was used.

Additional References for *In vitro* Bacterial Reverse Mutation Assay: None Found.

Type: ***In vitro* Clastogenicity Studies:** No Data.

Type: ***In vivo* Genetic Toxicity:** No Data.