WATER QUALITY CRITERIA FOR DIAZINON AND CHLORPYRIFOS

by

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SUMMARY

Recent toxicity information was reviewed and used to update freshwater and saltwater aquatic life criteria for diazinon and chlorpyrifos. These water quality criteria were compared to criteria developed by the U.S. Environmental Protection Agency (USEPA 1986;1998). The joint toxicity of diazinon and chlorpyrifos was also evaluated.

Thirteen new tests on the acute toxicity of diazinon to aquatic organisms were evaluated and 12 were accepted. These new values were pooled with values previously evaluated (Menconi and Cox 1994). The freshwater Final Acute Value (FAV) for diazinon was 0.16 µg/L. The freshwater Criterion Maximum Concentration (CMC) for diazinon was 0.08 µg/L. The draft CMC proposed by USEPA (1998) was 0.09 µg/L. No saltwater acute or chronic criteria were developed due to inadequate data. Six tests on the chronic toxicity of diazinon to aquatic organisms were evaluated and five were accepted. The freshwater Final Chronic Value (FCV) for diazinon was 0.05 µg/L. The freshwater Criterion Continuous Concentration (CCC) for diazinon was 0.05 µg/L. The USEPA (1998) did not propose a FCV or CCC for diazinon.

Twenty-five new tests on the acute toxicity of chlorpyrifos to aquatic organisms were evaluated and 13 were accepted. These new values were pooled with values previously evaluated (Menconi and Paul 1994). The freshwater FAV for chlorpyrifos was 0.05 µg/L. The freshwater CMC for chlorpyrifos was 0.02 µg/L. The freshwater CMC calculated by USEPA (1986) was 0.083 µg/L. The saltwater FAV for chlorpyrifos was 0.03 µg/L. The saltwater CMC was 0.02 µg/L. The saltwater CMC calculated by USEPA (1986) was 0.011 µg/L. One chronic toxicity test for chlorpyrifos was reviewed and accepted. The freshwater and saltwater FCVs for chlorpyrifos were 0.014 and 0.009 µg/L, respectively. The freshwater and saltwater CCCs for chlorpyrifos were 0.014 and 0.009 µg/L, respectively. Freshwater and saltwater CCCs calculated

by USEPA (1986) were 0.041 and 0.0056 μg/L, respectively.

Two studies on the joint toxicity of diazinon and chlorpyrifos to cladoceran *Ceriodaphnia dubia* were evaluated. Both studies suggest that the toxicities of diazinon and chlorpyrifos were additive.

TABLE OF CONTENTS

LIST OF TABLES

LIST OF ACRONYMS

ACR:	Acute-Chronic Ratio
ASTM:	American Society for Testing and Materials
CCC:	Criterion Continuous Concentration
CDFG:	California Department of Fish and Game
CDPR:	California Department of Pesticide Regulation
CMC:	Criterion Maximum Concentration
CVRWQCB:	Central Valley Regional Water Quality Control Board
FACR:	Final Acute-to-Chronic Ratio
FAV:	Final Acute Value
FCV:	Final Chronic Value
FPV:	Final Plant Value
FRV:	Final Residue Value
GMAV:	Genus Mean Acute Value
LOEC:	Lowest Observable Effect Concentration
MATC:	Maximum Acceptable Toxicant Concentration
NOEC:	No Observable Effect Concentration
SMAV:	Species Mean Acute Value
USEPA:	U.S. Environmental Protection Agency
WQC:	Water Quality Criterion

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INTRODUCTION

Several agencies (U.S. Geological Survey, Central Valley Regional Water Quality Control Board (CVRWQCB), and California Department of Pesticide Regulation (CDPR)) have detected diazinon and chlorpyrifos in the waters of the Sacramento-San Joaquin watershed beginning in the early 1990s. These detections have been the result of runoff from agricultural and urban areas (Domagalski et al. 1997; Kuivila and Foe 1995; Kratzer 1998; Ross et al. 1996). The California Department of Fish and Game (CDFG) had previously assessed the effects of diazinon and chlorpyrifos on aquatic organisms in the Sacramento-San Joaquin watershed (Menconi and Cox 1994; Menconi and Paul 1994). CDFG's hazard assessments are based on data from accepted tests and procedures outlined in U.S. Environmental Protection Agency (USEPA 1985) guidelines (Appendix A). However, data gaps were present which allowed only calculation of interim water quality criteria (WQC). In addition, more data were needed to evaluate the effect (i.e., antagonism, additivity, or synergism) of the joint action of diazinon and chlorpyrifos on aquatic organisms. This report gives an updated summary of the toxicity database of diazinon and chlorpyrifos, both alone and in mixtures, to aquatic organisms.

ACUTE TOXICITY OF DIAZINON TO AQUATIC ORGANISMS

Thirteen new tests on the acute toxicity of diazinon to aquatic organisms were evaluated (Appendix B). Twelve of these tests were found to be in general conformance with acceptability criteria developed by the USEPA (1985) and American Society for testing and Materials (ASTM 1996). Five of the ten accepted tests were on saltwater organisms. The remaining five accepted freshwater tests were used to revise WQC previously generated (Menconi and Cox 1994). For the previously generated acute WQC (Menconi and Cox 1994), fifty-nine tests were evaluated, and thirty-four tests were accepted (Appendix C). The Genus Mean Acute Values (GMAVs) for diazinon were calculated using all data; and these were ranked in ascending order (Table 1). The new freshwater toxicity tests evaluated for this report were for cladoceran *Ceriodaphnia dubia* (three tests), fathead minnow *Pimephales promelas*, and snail *Physa sp.* Data from eight freshwater families recommended by USEPA (1985) were available to derive a freshwater Final Acute Value (FAV) of 0.16 μg/L (Table 2). The freshwater FAV previously calculated using seven of the eight families was 0.16 μg/L (Menconi and Cox 1994). The freshwater FAV calculated by USEPA (1998) was 0.1826 μg/L.

Acceptable data were available for four of the eight saltwater families recommended by USEPA (1985) (Tables 3 and 4). A saltwater FAV was not calculated.

Table 1. Ranked Genus Mean Acute Values (GMAV) from accepted acute toxicity tests on freshwater species used to calculate the freshwater FAV for diazinon.

Rank	GMAV (µg/L)	Species	Number of tests
1	0.20	Amphipod	
		Gammarus fasciatus	1
2	0.44	Cladoceran	
		Ceriodaphnia dubia	7
3	1.06	Cladoceran, Genus *Daphnia*	
		Daphnia pulex (SMAV = 0.78)	3
		Daphnia magna (SMAV = 1.44)	2
4	1.59	Cladoceran	
		Simocephales serrulatus	2
5	4.15	Mysid	
		Neomysis mercedis	2
6	4.41	Snail	
		Physa sp.	1
7	25	Stonefly	
		Pteronarcys californica	1
8	272	Bluegill	
		Lepomis macrochirus	2
9	441	Salmonid, Genus *Oncorhynchus*	
		Oncorhynchus clarki (SMAV = 2166)	2
		Oncorhynchus mykiss (SMAV = 90)	3
10	660	Char, Genus *Salvelinus*	
		Salvelinus namaycush (SMAV = 602)	1
		Salvelinus fontinalis (SMAV = 723)	3
11	800	Guppy	
		Poecilia reticulata	1
12	1,643	Flagfish	
		Jordanella floridae	2
13	7,804	Fathead minnow	
		Pimephales promelas	5
14	8,000	Zebrafish	
		Brachydanio rerio	1
15	29,220	Rotifer	
		Brachionus calyciflorus	1

Table 2. Eight families of freshwater aquatic animals recommended by USEPA (1985) for use in deriving the Final Acute Value (FAV) and representative species for which diazinon acute toxicity data were available.

Family	Animal
1. One Salmonid	Brook trout
2. Another family in Osteichthyes	Bluegill
3. Another family in Chordata	Fathead minnow
4. One family not in Arthropoda or Chordata	Snail
5. One insect family or any phylum not already represented	Rotifer
6. One planktonic crustacean	Cladoceran
7. One benthic crustacean	Amphipod
8. One insect	Stonefly

Table 3. Ranked Genus Mean Acute Values (GMAV) from accepted acute toxicity tests on saltwater species for diazinon.

Rank	GMAV (µg/L)	Species	Number of tests
1	5.6	Mysid *Mysidopsis bahia*	3
2	21	Pink shrimp *Penaeus duorarum*	1
3	28	Rotifer *Brachionus plicatilis*	3
4	880	Eastern oyster *Crassostrea virginica*	1

Table 4. Eight families of saltwater aquatic animals recommended by USEPA (1985) for use in deriving the Final Acute Value (FAV) and representative species for which diazinon acute toxicity data were available.

Family	Animal
1, 2. Two families in chordata	N/A N/A
3. One family not in phylum Arthropoda or Chordata	Eastern oyster
4.5,6. Three other families not in phylum Chordata	Pink shrimp N/A N/A
7. A mysid or penaeid	Mysid
8. One other family not already represented	Rotifer

CHRONIC TOXICITY OF DIAZINON TO AQUATIC ORGANISMS

Six new chronic toxicity tests for diazinon were evaluated and five tests were accepted (Appendix B). The accepted tests were for cladoceran *Ceriodaphnia dubia*, sheepshead minnow *Cyprinodon variegatus*, mysid *Mysidopsis bahia*, and fathead minnow *Pimephales promelas* (two tests). An Acute-Chronic Ratio (ACR) value was generated for *C. dubia* using acute and chronic values from the same study (Table 5). There was no corresponding acute test for *C. variegatus*, so no ACR could be calculated. An ACR had previously been calculated for *M. bahia*, using acute and chronic values from the same study (Menconi and Cox 1994). For calculating the ACR, it is preferable to use acute and chronic values from the same study or at least the same laboratory. Therefore, the more recent chronic value for *M. bahia* was not used in the calculation of the ACR. An ACR had already been calculated for *P. promelas* (Menconi and Cox 1994). However, a new species mean ACR value was calculated using two sets of acute and chronic tests conducted in the same laboratory. The new species mean ACR for *P. promelas* was 196.

ACR values were calculated by dividing the FAV by the Maximum Acceptable Toxicant Concentration (MATC) for each species. Five ACR values were available for use in calculating the Final ACR (FACR) (Table 5). However, USEPA (1985) guidelines specify that if ACR values increase with increasing Species Mean Acute Values (SMAVs), only ACR values for those species with SMAVs close to the FAV should be used. It does appear that ACR values are lower for species acutely sensitive to diazinon. Therefore, only values for the three acutely sensitive species (*C. dubia, M. bahia,* and *D. magna*) were used in the calculation of the FACR. The calculated FACR was 3. The Final Chronic Value (FCV) is 0.05 µg/L. Most organophosphate insecticides have low FACR values based on the ACR values of acutely sensitive species. The FACR values for methyl parathion (Menconi and Harrington 1992) and chlorpyrifos (Menconi and Paul 1994) were 2.2 and 4, respectively.

Table 5. Available chronic tests and corresponding acute values and Acute-Chronic Ratio (ACR) values.

Species	Reference	MATC (µg/L)	LC_{50}(µg/L)	ACR
Cladoceran *Ceriodaphnia dubia*	Norberg-King (1987)	0.34	0.57	1.7
Cladoceran *Daphnia magna*	Surprenant (1988c)	0.23	1.44[a]	6.3
Fathead minnow *Pimephales promelas*	Norberg-King (1989)	25.0	9,350[b]	374
Fathead minnow *Pimephales promelas*	Jarvinen and Tanner (1982)	67	6,900[c]	103
Fathead minnow *Pimephales promelas*	Surprenant (1988d)	125	N/A[d]	N/A
Mysid *Mysidopsis bahia*	Nimmo et al. (1981)	1.9	4.82[c]	2.5
Mysid *Mysidopsis bahia*	Sousa et al. (1997a)	0.31	N/A	N/A
Sheepshead minnow *Cyprinodon variegatus*	Sousa et al. (1997b)	5.9	N/A	N/A

[a]Species Mean Acute Value: geometric mean of values from several tests on this species.
[b]Acute and chronic tests performed by the same laboratory.
[c]LC_{50} and MATC values from same test.
[d]Not Available. No corresponding acute value was available.

The USEPA (1998) did not calculate an FCV because they felt there was not a clear relationship between SMAVs and ACR values in their data set. When there is no trend apparent between SMAVs and ACR values and the ACR values are not within a factor of ten, USEPA (1985) guidelines specify that no chronic values can be calculated. Although there is overlap between the data sets for diazinon used by USEPA and CDFG, there are also several studies used in one report but not the other (Table 6). This difference is partially due to some studies being available to CDFG but not USEPA. Also, a few chronic studies that were accepted by the USEPA were rejected by DFG because the concentrations tested were inappropriate to generate Lowest Observable Effect Concentration (LOEC) and No Observable Effect Concentration (NOEC) values.

Table 6. Comparison of chronic toxicity tests for diazinon used by CDFG and USEPA

Reference	Organism	Used by USEPA?	Used by CDFG?	Comments
Allison (1977)	*Jordanella floridae*	Yes	No	Test rejected by CDFG because it did not generate an NOEC.
Allison and Hermanutz (1977)	*Salvelinus fontinalis*	Yes	No	Test rejected by CDFG (1994) because it did not generate an NOEC.
Goodman et al. (1979)	*Pimephales promelas*	Yes	No	Test rejected by CDFG (1994) because it did not generate an NOEC.
Jarvinen and Tanner (1982)	*Pimephales promelas*	Yes	Yes	
Nimmo et al. (1981)	*Mysidopsis bahia*	Yes	Yes	USEPA used original data to recalculate values; CDFG (1994) used values calculated by authors.
Norberg-King (1989)	*Pimephales promelas*	Yes	Yes	
Norberg-King (1987)	*Ceriodaphnia dubia*	Yes	No	Study not available to CDFG (cited in internal USEPA memo)
Surprenant (1988c)	*Daphnia magna*	No	Yes	Study not evaluated by USEPA, but accepted by CDFG (1994).

CRITERIA FOR DIAZINON

The freshwater FAV for diazinon was 0.16 µg/L. The FACR for diazinon was 3. The FCV for diazinon was 0.05 µg/L . The USEPA guidelines specify that a WQC consists of two concentrations, the Criterion Maximum Concentration (CMC) and the Criterion Continuous Concentration (CCC). The CMC was equal to one-half the FAV or 0.08 µg/L (Table 7). Freshwater organisms should not be affected unacceptably if the one-hour average concentration of diazinon does not exceed 0.08 µg/L more than once every three years on the average. The CCC is equal to the lowest of three values: the FCV, the Final Plant Value (FPV), of the Final Residue Value (FRV). Diazinon does not appear to bioconcentrate to a significant degree (Kanazawa 1978), and diazinon is more toxic to animals than to plants. Therefore, the CCC was equal to the FCV of 0.05 µg/L. Freshwater organisms should not be affected unacceptably if the four-day average concentration for diazinon does not exceed 0.04 µg/L more than once every three years on average. WQC are for diazinon alone.

Table 7. CDFG and USEPA (1998) water quality criteria for diazinon to freshwater organisms.

Reference	CMC	CCC
CDFG (this report)	0.08 µg/L	0.05 µg/L
USEPA (1998)	0.09 µg/L	not calculated

ACUTE TOXICITY OF CHLORPYRIFOS TO AQUATIC ORGANISMS

Twenty-five new tests on the acute toxicity of chlorpyrifos to aquatic organisms were evaluated (Appendix B). Thirteen of these tests were found to be in general conformance with acceptability criteria adapted from the USEPA (1985) and ASTM (1996). Three of the thirteen accepted tests were on saltwater organisms. The remaining ten accepted freshwater tests were used to revise freshwater WQC previously generated (Menconi and Paul 1994). For the previously generated acute WQC (Menconi and Paul 1994), one hundred and nine tests were evaluated, and seventy were accepted (Appendix D). GMAVs for chlorpyrifos were calculated using data from Menconi and Paul (1994) and more recent data; and these are ranked in ascending order (Table 8). The new freshwater toxicity tests evaluated for this report were for cladocerans *Ceriodaphnia dubia* (3 tests) and *Daphnia pulex*, amphipod *Hyalella azteca*, fathead minnow *Pimephales promelas* (2 tests), and midge *Chironomus tentans* (3 tests). Data from all eight freshwater families recommended by USEPA (1985) were available to derive a freshwater FAV (Table 9). The calculated freshwater FAV was 0.05 µg/L. The freshwater FAV previously calculated was 0.07 µg/L (Menconi and Paul 1994). The freshwater FAV calculated by USEPA (1986) was 0.1669 µg/L. The freshwater FAV calculated by Menconi and Paul (1994) and this report used toxicity values for sensitive species not available for use in the USEPA (1986) criteria.

GMAVs for saltwater organisms were calculated using all data and ranked in ascending order (Table 10). Acceptable data were available for all eight saltwater taxa (Table 11). The calculated saltwater FAV was 0.03 µg/L. This value is the same as the previously calculated FAV (Menconi and Paul 1994). The saltwater FAV calculated by USEPA (1986) was 0.02284 µg/L.

Table 8. Ranked Genus Mean Acute Values (GMAV) from accepted acute toxicity tests on freshwater species used to calculate the freshwater FAV for chlorpyrifos.

Rank	GMAV (µg/L)	Species	Number of tests
1	0.06	Cladoceran	
		Ceriodaphnia dubia	5
2	0.11	Amphipod	
		Gammarus lacustris	1
3	0.15	Mysid	
		Neomysis mercedis	3
4	0.38	Stonefly	
		Pteronarcella badia	1
5	0.54	Cladoceran, Genus *Daphnia*	
		Daphnia pulex (SMAV = 0.30)	1
		Daphnia magna (SMAV = 1.0)	1
6	0.58	Stonefly	
		Claassenia sabulosa	1
7	0.60	Midge	
		Chironomus tentans	3
8	0.80	Crawling water beetle	
		Petodytes sp.	1
9	3.03	Bluegill	
		Lepomis macrochirus	6
10	6.0	Crayfish	
		Orconectes immunis	1
11	10	Stonefly	
		Pteronarcys californica	1
12	10.1	Salmonid, Genus *Oncorhynchus*	
		Oncorhynchus mykiss (SMAV = 7.5)	3
		Onchorynchus clarki (SMAV = 13.6)	4
13	138	Amphipod	
		Hyallela azteca	1
14	244	Lake trout	
		Salvelinus namaycush	1
15	274	Fathead minnow	
		Pimephales promelas	5
16	475	Channel catfish	
		Ictalurus punctatus	2
17	>806	Goldfish	
		Carassius auratus	1
18	>806	Snail	
		Aplexa hypnorum	1

Table 9. Eight families of freshwater aquatic animals recommended by USEPA (1985) for use in deriving the Final Acute Value (FAV) and representative species for which chlorpyrifos acute toxicity data were available.

Family	Animal
1. One Salmonid	Rainbow trout
2. Another family in Osteichthyes	Bluegill
3. Another family in Chordata	Fathead minnow
4. One family not in Arthropoda or Chordata	Amphipod
5. One insect family or any phylum not already represented	Stonefly
6. One planktonic crustacean	Cladoceran
7. One benthic crustacean	Crayfish
8. One insect	Crawling water beetle

Table 10. Ranked Genus Mean Acute Values (GMAV) from accepted acute toxicity tests on saltwater species used to calculate the saltwater FAV for chlorpyrifos.

Rank	GMAV (µg/L)	Species	Number of tests
1	0.04	Mysid	
		Mysidopsis bahia	3
2	0.69	Shrimp, Genus *Penaeus*	
		Penaeus aztecus (SMAV = 0.20)	1
		Penaeus duorarum (SMAV = 2.4)	1
3	1.2	California grunion	
		Leuresthes tenuis	6
4	1.5	Grass shrimp	
		Palaemonetes pugio	1
5	1.5	Silverside, Genus *Menidia*	
		Menidia menidia (SMAV = 0.5)	5
		Menidia peninsiluae (SMAV = 1.6)	9
		Menidia beryllina (SMAV = 4.2)	1
6	1.6	Rotifer	
		Brachionus plicatilis	3
7	2.7	Killifish, Genus *Fundulus*	
		Fundulus grandis (SMAV = 1.8)	1
		Fundulus similis (SMAV = 4.1)	1
8	5.2	Blue crab	
		Callinectes sapidus	1
9	5.4	Striped mullet	
		Mugil cephalus	1
10	7.0	Spot	
		Leiostomus xanthurus	1
11	188	Gulf toadfish	
		Opsanus beta	2
12	194	Sheepshead minnow	
		Cyprinodon variegatus	2
13	1991	Easter oyster	
		Crassostrea virginica	1

Table 11. Eight families of saltwater aquatic animals recommended by USEPA (1985) for use in deriving the Final Acute Value (FAV) and representative species for which chlorpyrifos acute toxicity data were available.

Family	Animal
1, 2. Two families in chordata	Silverside
	Striped mullet
3. One family not in phylum Arthropoda or Chordata	Eastern oyster
4.5,6. Three other families not in phylum Chordata	Brown shrimp
	Blue crab
	Pink shrimp
7. A mysid or penaeid	Mysid
8. One other family not already represented	Rotifer

CHRONIC TOXICITY OF CHLORPYRIFOS TO AQUATIC ORGANISMS

One chronic toxicity test for chlorpyrifos was evaluated and accepted. This test was for cladoceran *Ceriodaphnia dubia*. As a part of the same study, an acute test was conducted for *C. dubia*. The ACR generated in this study was added to the ACRs previously generated (Menconi and Paul 1994) and a new FACR was calculated (Table 12). USEPA (1985) guidelines specify that if the ACR values increase or decrease as the SMAVs increase, as the ACR values generally do with chlorpyrifos, only species with SMAVs close to the FAV should be used to calculate the FACR. Accordingly, the FACR value for chlorpyrifos was calculated as the geometric mean of the ACR values for cladoceran *Ceriodaphnia dubia* and mysid *Mysidopsis bahia*. The FACR was 3.5. The freshwater FCV was 0.014 μg/L and the saltwater FCV was 0.009 μg/L. These Freshwater and saltwater FCVs generated by Menconi and Paul (1994) were 0.02 and 0.01 μg/L, respectively. The freshwater and saltwater FCVs generated by USEPA (1986) were 0.04107 and 0.005620 μg/L, respectively.

Table 12. Acute-Chronic Ratio (ACR) values for freshwater and saltwater species for chlorpyrifos for which acute and chronic toxicity data were available.

Species	MATC (µg/L)	LC_{50}(µg/L)	ACR
Cladoceran *Ceriodaphnia dubia*	0.040	0.038[a]	0.95[c]
Tidewater silverside *Menidia peninsulae*	0.54	0.71[b]	1.3
Inland silverside *Menidia beryllina*	1.16	4.2	3.6
Mysid *Mysidopsis bahia*	0.003	0.040[b]	13.3[c]
Fathead minnow *Pimephales promelas*	5.23	249[a]	47.6
Fathead minnow *Pimephales promelas*	2.26	140[a]	61.9
Sheepshead minnow *Cyprinodon variegatus*	2.26	194[b]	85.8
Gulf toadfish *Opsanus beta*	2.28	520[a]	228

[a]LC_{50} and MATC from same test.
[b]Species Mean Acute Value: geometric mean of values from several tests on this species.
[c]ACR value used to calculate Final ACR value.

CRITERIA FOR CHLORPYRIFOS

The freshwater and saltwater FAVs for chlorpyrifos were 0.05 µg/L and 0.03 µg/L, respectively. The FACR for chlorpyrifos was 3.5. The freshwater and saltwater FCVs for chlorpyrifos were 0.014 and 0.009 µg/L, respectively . The freshwater CMC was equal to one-half the freshwater FAV, or 0.02 µg/L The saltwater CMC was equal to one-half the saltwater FAV, or 0.02 µg/L Freshwater and saltwater organisms should not be affected unacceptably if the one-hour average concentration of chlorpyrifos does not exceed 0.02 µg/L more than once every three years on average. The CCC is equal to the lowest of three values: the FCV, the FPV, or the FRV. Therefore, the freshwater and saltwater CCC values were 0.014 and 0.009 µg/L, respectively. Freshwater and saltwater organisms should not be affected unacceptably if the four-day concentration of chlorpyrifos does not exceed 0.014 µg/L and 0.009 µg/L, respectively, more than once every three years on average. The freshwater CMC and CCC generated by USEPA (1986) were 0.083 and 0.041 µg/L, respectively (Table 12). The saltwater CMC and CCC calculated by USEPA (1986) was 0.011 µg/L and 0.0056 µg/L, respectively. These WQC are for chlorpyrifos alone.

Table 13. CDFG and USEPA (1986) water quality criteria for chlorpyrifos to freshwater organisms

Reference	CMC	CCC
CDFG (this report)	0.02 µg/L	0.014 µg/L
USEPA (1986)	0.083 µg/L	0.041 µg/L

JOINT TOXICITY OF DIAZINON AND CHLORPYRIFOS

Two studies were conducted to evaluate the joint toxicity of diazinon and chlorpyrifos to the cladoceran *Ceriodaphnia dubia* (Bailey et al. 1997, CDFG 1999a). The toxicities of chlorpyrifos and diazinon appear additive (Table 14). An Additive Index (Marking 1985) between -1 and 1 (symmetrical about 0) indicates additivity (Table 14).

Table 14. Joint toxicity of diazinon and chlorpyrifos (96-h LC_{50} values in µg/L) to *Ceriodaphnia dubia*.

	Bailey et al. (1997)	CDFG (1999a,c; 1998b)
Chlorpyrifos alone	0.053, 0.055	0.038
Diazinon alone	0.32, 0.35	0.44
Chlorpyrifos in mixture	0.024, 0.020 (0.41 toxic unit)	0.02 (0.52 toxic unit)
Diazinon in mixture	0.23, 0.24 (0.70 toxic unit)	0.15 (0.34 toxic unit)
Total Toxic Units	1.11	0.88
Additive Index	-0.11	0.14

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APPENDIX A. Procedure Used By the California Department of Fish and Game to Prepare Hazard Assessments.

The California Department of Fish and Game (CDFG) Pesticides Investigations Unit assesses the hazard of pesticides to aquatic organisms. The hazard assessment procedure includes evaluation of toxicity studies, establishment of the Water Quality Criterion (WQC) and assessment of potential hazards.

Acute and chronic toxicity data are obtained from studies published in scientific literature and laboratory reports required by the U.S. Environmental Protection Agency for pesticide registration. The CDFG evaluates the quality of these data by evaluating the tests for compliance with standards (ASTM 1988, 1992) for test type, method, design, species and for water quality standards and toxicant monitoring and maintenance. Although a study need not comply with every standard, tests are rejected if they do not observe certain fundamental procedures or if several important standards are not met. Studies are also rejected if they do not contain sufficient information to be properly evaluated and the necessary information cannot be obtained from the researcher.

Acute toxicity data from acceptable tests on freshwater and saltwater organisms are used to determine a Final Acute Value (FAV). The USEPA (1985) guidelines recommend eight categories of freshwater and saltwater organisms for deriving freshwater and saltwater FAVs. Generally, these categories of organisms are available, commonly used, and testing procedures are outlined

The FAV is calculated as follows:

10 Species Mean Acute Values (SMAV) are calculated as the geometric mean of LC_{50} and EC_{50} values from all accepted toxicity tests performed on that species.

20 Genus Mean Acute Values (GMAV) are calculated as the geometric mean of all SMAVs for each genus.

30 The GMAVs are ranked (R) from “1” for the lowest to “N” for the highest. Identical GMAVs are arbitrarily assigned successive ranks.

4. The cumulative probability (P) is calculated for each GMAV as R/ (N+1).

50 The four GMAVs with cumulative probabilities closest to 0.05 are selected. If fewer than 59 GMAVs are available, these will always be the four lowest GMAVs.

60 The FAV is calculated using the selected GMAVs and cumulative probabilities (P), as follows:

$$S^2 = \frac{E\ [\ (\ln GMAV)^2\ ] - [\ (E(\ln GMAV)\ )^2]/4}{E(P) - [\ (\ E(\_P)\ )^2]/4}$$

$L = [\,E\ (\ln GMAV) - S\ (E\ (\_P)\ )^2]/4$
$A = S\ (\_0.05) + L$

$FAV = e^A$

Chronic toxicity data from acceptable tests on freshwater and saltwater organisms are used to determine a Final Chronic Value (FCV). If data are available for the eight families, the FCV is calculated using the same procedure as described for the FAV. If sufficient data are not available, the following procedure is used:

1. Chronic values are obtained by calculating the geometric mean of the NOEC and the LOEC values from accepted chronic toxicity tests.

2. Acute-Chronic ratios (ACR) are calculated for each chronic value for which at least one corresponding acute value is available. Whenever possible, the acute test (s) should be part of the same study as the chronic test.

3. The Final ACR (FACR) is calculated as the geometric mean of all mean ACRs available for both freshwater and saltwater species.

4. FCV = FAV / FACR.

Plant toxicity data from algae or aquatic vascular plants are used to determine a Final Plant Value (FPV). The FPV is the lowest result from a test with a biologically important endpoint.

The USEPA guidelines specify that a WQC consists of two concentrations, the Criterion Maximum Concentration (CMC) and the Criterion Continuous Concentration (CCC). The CMC is equal to one-half the FAV. The CCC is equal to the lowest of three values: the FCV, the FPV, or the Final Residue Value (FRV). The FRV is intended to prevent pesticide concentrations in recreational or commercially important species from affecting marketability because of excedence of applicable action levels and to protect important resident species (USEPA 1985).

The WQC is stated as follows: (freshwater / saltwater) aquatic organisms should not be affected unacceptably if the four-day average concentration of (pesticide) does not exceed (CCC value) and if the one-hour average concentration does not exceed (CMC value) more than once every three years on the average (USEPA 1985).

Hazard assessment is an iterative process by which new data are evaluated to refine the WQC. Hazard assessments frequently recommend additional toxicity tests with sensitive native species and commonly used test organisms listed by ASTM.

APPENDIX B. Abstracts of Accepted and Unaccepted Acute and Chronic Toxicity Tests Reviewed for Hazard Assessment.

Accepted acute toxicity tests- The following tests used accepted test methods.

Bailey et al. (1997) - In 1996, 48- and 96-h acute static toxicity tests were conducted by the U.C. Davis Aquatic Toxicology Laboratory and AQUA-Science Laboratory in Davis, California with mixtures of diazinon (99%) and chlorpyrifos (99%) on <24-h cladoceran *Ceriodaphnia dubia*. Five combinations of concentrations were tested with four replicates and water controls were used. Water quality parameters during the tests were: temperature of 24 to 25°C; dissolved oxygen of 7.6 to 8.4 mg/L; pH of 7.40 to 8.23; and hardness of 80 to 100 mg/L as $CaCO_3$. Control survival was acceptable. The 96-h LC_{50} values were 0.32 and 0.35 μg/L for diazinon and 0.053 and 0.055 μg/L for chlorpyrifos. Based on the ratios of concentrations tested, the 96-h LC_{50} values for the mixture tests were 0.23 and 0.24 μg/L for diazinon and 0.020 and 0.024 μg/L for chlorpyrifos.

Brown et al. (1997) - In 1997, a 96-h acute static toxicity test was conducted by the Dow Chemical Company Toxicology Research Laboratory in Midland, Michigan with chlorpyrifos (98%) on juvenile amphipod *Hyalella azteca*. Six concentrations and a control were tested in replicate. Water quality parameters during the test were: temperature of 18 $\pm$ 1°C; dissolved oxygen of >93% saturation; pH of 7.1 to 7.8; and hardness of 166 mg/L as $CaCO_3$. Control survival was at least 90% and mortality range was acceptable. The 96-h EC_{50} value for chlorpyrifos was 138 μg/L.

CDFG (1999a) - In 1999, a 96-h acute static toxicity test was conducted by the California Department of Fish and Game Aquatic Toxicology Laboratory in Elk Grove, California with a mixture of diazinon (87%) and chlorpyrifos (99.8%) on <24-h cladoceran *Ceriodaphnia dubia*. Five concentrations and water and solvent controls were tested. Water quality parameters during the test were: temperature of 24.2 to 25.6°C; pH of 8.03 to 8.60; dissolved oxygen of 4.50 to 8.20 mg/L; and hardness of 176 to 178 mg/L as $CaCO_3$. Control survival was 100% and mortality range was acceptable. Based on the ratio of concentrations tested, the 96-h LC_{50} values were 0.02 μg/L for chlorpyrifos and 0.15 μg/L for diazinon.

CDFG (1999c) - In 1999, a 96-h acute static toxicity test was conducted by the California Department of Fish and Game Aquatic Toxicology Laboratory in Elk Grove, California with chlorpyrifos (99.8%) on <24-h cladoceran *Ceriodaphnia dubia*. Five concentrations and water and solvent controls were tested. Water quality parameters during the test were: temperature of 24.0 to 25.2°C; pH of 7.45 to 8.47; dissolved oxygen of 7.45 to 9.10 mg/L; and hardness of 176 to 178 mg/L as $CaCO_3$. Control survival was 100% and mortality range was acceptable. The 96-h LC_{50} for chlorpyrifos was 0.038 μg/L.

CDFG (1998a) - In 1998, a 96-h acute static toxicity test was conducted by the California Department of Fish and Game Aquatic Toxicology Laboratory in Elk Grove, California with diazinon (87%) on juvenile snail *Physa sp.*. Five concentrations and a water control were tested. Water quality parameters during the test were: temperature of 21.0 to 22.4^oC; pH of 6.72 to 8.18; dissolved oxygen of 4.12 to 8.81 mg/L; and hardness of 110 mg/L as $CaCO_3$. Control survival was 95% and mortality range was acceptable. The 96-h LC_{50} for diazinon was 4.41 μg/L.

CDFG (1998b) - In 1998, a 96-h acute static toxicity test was conducted by the California Department of Fish and Game Aquatic Toxicology Laboratory in Elk Grove, California with diazinon (87.3%) on <24-h cladoceran *Ceriodaphnia dubia*. Five concentrations and a water control were tested. Water quality parameters during the test were: temperature of 24.2 to 25.2^oC; pH of 7.62 to 8.19; dissolved oxygen of 6.23 to 8.37 mg/L; and hardness of 116 mg/L as $CaCO_3$. Control survival was 100% and mortality range was acceptable. The 96-h LC_{50} for diazinon was 0.436 μg/L.

Cripe (1994) - In 1994, 96-h acute static toxicity tests were conducted by the U.S. Environmental Protection Agency in Gulf Breeze, Florida with diazinon (99%) on <24-h mysid *Mysidopsis bahia* and 3 to 5-d pink shrimp *Penaeus duorarum*. Five concentrations were tested in replicate with seawater and solvent controls. Water quality parameters during the tests were: temperature of 25 + 0.5^oC; average dissolved oxygen of 5.9 mg/L for mysid and 5.6 mg/L for shrimp; pH of 7.5 to 8.1; and salinity of $25^o/_{oo}$. Control survival was acceptable. The 96-h LC_{50} values for diazinon were 8.5 μg/L for mysid and 21 μg/L for shrimp.

Geiger et al. (1988) - From 1979 to 1985, three 96-h acute flow-through toxicity tests were conducted by the USEPA Environmental Research Laboratory in Duluth, Minnesota with diazinon (87.1%) and chlorpyrifos (99.9%) (two tests) on 27-44 day-old fathead minnows *Pimephales promelas*. Five concentrations were tested with water controls. Water quality parameters during the test were: temperature of 25.5^oC for diazinon and 25.1 and 16.3^oC for chlorpyrifos; dissolved oxygen of 6.6 mg/L for diazinon and 8.1 and 7.6 mg/L for chlorpyrifos; pH of 7.6 for diazinon and 7.5 and 7.6 for chlorpyrifos; and hardness of 43.6 mg/L as $CaCO_3$ for diazinon and 44.4 and 46.5 mg/L as $CaCO_3$ for chlorpyrifos. Control survival was 100% for all tests. The 96-h LC_{50} values were 9,350 μg/L for diazinon and 200 and 506 μg/L for chlorpyrifos.

Guzzella et al. (1997) - In 1996, 24-h acute static toxicity tests were conducted by the Water Research Institute in Milan, Italy with diazinon (>95%) and chlorpyrifos (>95%) on neonates of rotifer *Brachionus plicatilis*. Five concentrations with five replicates and water controls were tested. The temperature during the test was 25^oC; no other water quality parameters were given. Control survival was acceptable. The 24-h LC_{50} values were 30, 27, and 27 μg/L for diazinon and 1.4, 1.9, and 1.7 μg/L for chlorpyrifos.

Jarvinen and Tanner (1982) - In 1982, a 96-h acute flow-through toxicity test was conducted by the USEPA Environmental Research Laboratory in Duluth, Minnesota with diazinon (87%) on larval fathead minnow *Pimephales promelas*. Five concentrations were tested in replicate with water controls. Water quality parameters during the test were: temperature of 23.5 to 26^{o}C;

dissolved oxygen of 6.5 to 8.4 mg/L; pH of 7.4 to 7.8; and hardness of 45.8 mg/L as $CaCO_3$. Control survival was acceptable. The 96-h LC_{50} value for diazinon was 6,900 μg/L.

Norberg-King (1987) - In 1987, a 48-h acute static toxicity test was conducted by USEPA Environmental Research Laboratory in Duluth, Minnesota with diazinon (85%) on <24-h cladoceran *Ceriodaphnia dubia*. Water quality parameters during the test were acceptable. Control survival during the test was 100%. The 48-h LC_{50} value for diazinon was 0.57 mg/L.

Pape-Lindstrom and Lydy (1997) - In 1996, 96-h acute static toxicity tests were conducted by Wichita State University in Wichita, Kansas with chlorpyrifos (99%) on fourth instar midge *Chironomus tentans*. Five concentrations with replicates and solvent and water controls were tested. Water quality parameters during the tests averaged: temperature of 20^{o}C; dissolved oxygen of 88.8% saturation; and pH of 7.95. Control survival was acceptable. The 96-h EC_{50} values, based on immobilization, for chlorpyrifos were 0.51, 0.58, and 0.75 μg/L.

Van Der Hoeven and Gerritsen (1997) - In 1996, a 48-h acute static toxicity test was conducted by TNO-Environmental Sciences in the Netherlands with technical grade chlorpyrifos on <24-h cladoceran *Daphnia pulex.* Five concentrations were tested with controls. Water quality parameters during the tests were: temperature of 20 $\pm$ 1^{o}C; pH of 8.0 to 8.2; and hardness of 220 mg/L as $CaCO_3$. The 48-h LC_{50} value for chlorpyrifos was 0.30 μg/L.

Unaccepted acute toxicity tests - The following tests did not use accepted test methods and/or produce acceptable results.

Ali et al. (1995) - In 1995, 24-h acute static toxicity tests were conducted by the University of Florida in Sanford, Florida with chlorpyrifos (99%) on third and fourth instar mosquito *Aedes albopictus*. Four to nine concentrations were tested in replicate with solvent controls. Water quality parameters were not given. Control survival and mortality range were not given. The 24-h LC_{50} for chlorpyrifos was 0.0033 mg/L. This value was not used because information regarding mortality range and control mortality was not given.

Olima et al. (1997) - In 1997, nine 96-h acute static toxicity tests were conducted by the University of Technology in Sydney, Australia with chlorpyrifos on freshwater shrimp *Paratya australiensis*. Five to six concentrations and solvent controls were tested in triplicate. Water quality parameters during the test were: temperature of 23.0 $\pm$ 1^{o}C; dissolved oxygen of >75% saturation; and pH of 6.70 to 7.50. Control survival and mortality range were not given. The 96-h LC_{50} for chlorpyrifos was 0.08 to 0.28 µg/L. These values were not used because the shrimp were field collected and may have already been exposed to chlorpyrifos.

Rice et al. (1997) - In 1996, a 48-h acute static toxicity test was conducted by the Iowa State Pesticide Toxicology Laboratory in Ames, Iowa with chlorpyrifos (99%) on freshwater fish *Oryzias latipes*. Five concentrations and a solvent control were tested in triplicate. Water quality parameters during the test were: temperature of 25 $\pm$ 1^{o}C; dissolved oxygen of 7.1 $\pm$ 1.3 mg/L; pH of 7.3 $\pm$ 0.7; and hardness of 136 $\pm$ 20 mg/L as $CaCO_3$. Control survival and mortality range were not given. The 48-h LC_{50} for chlorpyrifos was 250 µg/L. This value was not used because the species tested is not resident in North America.

Van Wijngaarden et al. (1996) - In 1990, 48-h acute toxicity tests were conducted by the DLO Winand Staring Centre for Integrated Land, Soil, and Water Research in Wageningen, the Netherlands with Dursban 4E (45% chlorpyrifos) on several aquatic invertebrates. Water quality parameters and control survival was not given. The values generated by this test were not used because the percent of active ingredient in the formulated product was too low and essential information was not given.

Villar et al. (1994) - In 1992, 96-h acute static toxicity tests were conducted by the University of Cordoba in Spain with diazinon (38%) on planarian *Dugesia tigrina*. The number of concentrations and controls tested and water quality parameters were not given. The 96-h LC_{50} value for diazinon was 630 µg/L. This value was not used because the percent of active ingredient in the formulated product was too low and essential information was not given.

Accepted chronic toxicity tests - The following tests used accepted test methods.

CDFG (1999b) - In 1999, a static chronic toxicity test was conducted by the California Department of Fish and Game Aquatic Toxicology Laboratory in Elk Grove, California with chlorpyrifos (99.9%) on <24-h cladoceran *Ceriodaphnia dubia*. Five concentrations were tested with water and solvent controls. Water quality parameters during the test were: temperature of 24.0 to 25.1^{o}C; pH of 7.90 to 8.58; hardness of 168 to 188 mg/L as $CaCO_3$; and dissolved oxygen of 3.63 to 9.88 mg/L. Control survival during the test was 80 to 100%. The 7-d NOEC, LOEC, and MATC values were 0.029, 0.054, and 0.040 µg/L, respectively.

Jarvinen and Tanner (1982) - In 1982, a 32-d flow-through chronic toxicity test was performed by the USEPA Environmental Research Laboratory in Duluth, Minnesota with diazinon (87%) on <48-h embryos of fathead minnow *Pimephales promelas.* Five concentrations were tested in replicate with a dilution water control. Water quality parameters during the test were: temperature of 25.0 $\pm$ 0.6^{o}C; pH of 7.4 to 7.8; dissolved oxygen of 6.5 to 8.4 mg/L; and hardness of 45.8 mg/L as $CaCO_3$. Control survival was acceptable. The NOEC, LOEC, and MATC values were 0.050, 0.090, and 0.067 µg/L, respectively.

Norberg-King (1989) - In 1988, 7-d and 32-d chronic renewal and flow-through tests were conducted by the USEPA Environmental Research Laboratory in Duluth, Minnesota with diazinon (88.2%) on larval fathead minnows *Pimephales promelas.* Five concentrations with four replicates and water controls were used. Water quality parameters were acceptable. Control survival was acceptable. The NOEC and LOEC values for the 32-d test was 16.5 and 37.8 µg/L, respectively. Values from the 7-d tests were considerably higher and were not used.

Norberg-King (1987) - In 1987, a 7-d chronic renewal test was conducted by the USEPA Environmental Research Laboratory in Duluth, Minnesota with diazinon (85%) on <6-h cladoceran *Ceriodaphnia dubia*. Five concentrations were tested with ten replicates and dilution water controls. Water quality parameters were acceptable. Control survival was 100%. The NOEC, LOEC, and MATC values were 0.220, 0.520, and 0.34 µg/L, respectively.

Sousa (1997a) - In 1997, a flow-through chronic toxicity test was conducted by Springborn Life Sciences Laboratory in Wareham, Massachusetts with diazinon (87.3%) on <24-h mysid *Mysidopsis bahia*. Five concentrations and solvent and water controls were tested. Water quality parameters during the test were: temperature of 24 to 25^{o}C; pH of 7.7 to 8.3; dissolved oxygen of 68 to 100% saturation; and salinity of 25 to $26^{o}/_{oo}$. Control survival was 90 to 92%. The 28-d MATC for diazinon was 0.31 µg/L.

Sousa (1997b) - In 1997, a flow-through chronic toxicity test was conducted by Springborn Life Sciences Laboratory in Wareham, Massachusetts with diazinon (87.3%) on <24-h sheepshead minnow *Cyprinodon variegatus*. Five concentrations and solvent and water controls were tested. Water quality parameters during the test were: temperature of 23 to 26°C; pH of 7.4 to 7.9;

dissolved oxygen of 5.8 to 7.6 mg/L; and salinity of 30 to $32^{o}/_{oo}$. Control survival was 95 to 100%. The 34-d MATC for diazinon was 5.9 µg/L.

Unaccepted chronic toxicity tests - The following tests did not use accepted test methods and/or produce acceptable results.

Allison (1977) - In 1977, a life-cycle chronic test was conducted by the USEPA Environmental Research Laboratory at Duluth, Minnesota with technical grade diazinon (92.5%) on flagfish *Jordanella floridae*. Five concentrations and solvent and water controls were tested. Chronic effects were observed at all concentrations. The results from this test were not used because no NOEC was generated.

APPENDIX C. Acute and Chronic Tests Evaluated for Diazinon in Menconi and Cox (1994).

Accepted acute toxicity tests - The following tests for diazinon evaluated in Menconi and Cox (1994) used accepted test methods.

Allison and Hermanutz (1977), Hermanutz (pers. comm.) - From 1971 to 1973, 96-h flow-through toxicity tests were performed by the EPA on technical grade diazinon (92.5%) with 13-, 15-, and 20- week old fathead minnow *Pimephales promelas* (three tests), 1-year old bluegill *Lepomis macrochirus* (two tests), 1-y old brook trout *Salvelinus fontinalis* (three tests), and 6 week old flagfish *Jordanella floridae* (two tests). American Public Health Association (APHA) (1971) test standards were used. Five concentrations were tested in replicate and a water control was included. Concentrations were measured three to six times during the test. Water quality parameters during the tests averaged: temperature 25 $\pm$1^{o}C (fathead minnow), 25 $\pm$0.5^{o}C (bluegill), 12 $\pm$0.5^{o}C (brook trout), and 25 $\pm$0.5^{o}C (flagfish); pH 7.3-7.5; dissolved oxygen 65-105% saturation; and hardness 44-45 mg/L as $CaCO_3$. Control survival was not mentioned. The 96-h LC_{50} values were: fathead minnow: 10,000 µg/L, 6,800 µg/L and 6,600 µg/L ; bluegill: 440 µg/L; brook trout: 800 µg/L, 450 µg/L , and 1,050 µg/L; flagfish: 1,500 µg/L and 1,800 µg/L. One test performed with bluegill was not included because an insufficient number of organisms was used (ASTM 1988a). Chronic tests were also conducted with fathead minnow and brook trout.

Ankley et al. (1991), Ankley (pers. comm.) - In 1991, 48-h static toxicity tests were performed by the EPA and AScI Corporation on technical grade diazinon (95-99%) with $\leq$48-h old cladocerans *Ceriodaphnia dubia*, *Daphnia magna* and *Daphnia pulex*. EPA (1990) test standards were used. Five concentrations were tested in replicate and water controls were used. Concentrations were not measured during the test. Water quality parameters during the test averaged: temperature of 25°C; pH of 7.6-8.0; dissolved oxygen of $\geq$90% saturation; and hardness of 160-180 mg/L. Control survival was $\geq$90%. The 48-h LC_{50} values were: *Ceriodaphnia dubia*: 0.50 µg/L; *Daphnia magna*: 0.80 µg/L and *Daphnia pulex*: 0.65 µg/L.

CDFG (1992a) - In 1992, 96-h static renewal toxicity tests were performed by the CDFG Aquatic Toxicology Laboratory on technical grade diazinon (87%) with neonate cladoceran *Ceriodaphnia dubia*. EPA (1990), ASTM (1988a) test standards were used. Five concentrations were tested and water and solvent controls were used. Concentrations were measured during the test. Water quality parameters during the test averaged: temperature of 24.4°C; pH of 8.3; hardness of 123.5 mg/L; conductivity of 382.5 µs/cm; salinity not mentioned; alkalinity of 112.0 mg/L and dissolved oxygen of 8.0 mg/L. Water control survival was 90% and solvent control survival was 100%. The 96-h LC_{50} value was 0.47 µg/L, the No Observable Effect Concentration (NOEC) and the Lowest Observable Effects Concentration (LOEC) values based on immobilization were 0.354 µg/L and 0.625 µg/L, respectively.

CDFG (1992b) - In 1992, a 96-h static renewal toxicity test was performed by the CDFG Aquatic Toxicology Laboratory on technical grade diazinon (87%) with neonate mysids *Neomysis mercedis*. ASTM (1992) test standards were used. Five concentrations of diazinon were tested and solvent and water controls were used. Concentrations were measured during the test. Water quality parameters during the test averaged: temperature of 17°C; pH of 8.3; hardness of 457.0 mg/L; salinity of 1.8 $^{o}/_{oo}$; conductivity of 3002.78 µs/cm; alkalinity of 149.63 mg/L; and dissolved oxygen of 8.71 mg/L. Control survival was 100%. The 96-h LC_{50} value was 3.57 µg/L, the NOEC and LOEC values, based on immobilization, were 2.10 µg/L and 4.15 µg/L, respectively.

CDFG (1992c) - In 1992, a 96-h static renewal test was performed by the CDFG Aquatic Toxicology Laboratory on technical grade diazinon (87%) with neonate cladoceran *Ceriodaphnia dubia*. EPA (1990), ASTM (1988a) test standards were used. Five concentrations were tested and water and solvent controls were used. Concentrations were measured during the test. Water quality parameters during the test averaged: temperature of 24.4°C; pH of 8.5; hardness of 125.0 mg/L $CaCo_3$; conductivity of 388.7 µs/cm; alkalinity of 100.0 mg/L; dissolved oxygen of 7.81 mg/L. Control survival was 100%. The 96-h LC_{50} value was 0.507 µg/L, the NOEC and LOEC values based on immobilization were 0.345 µg/L and 0.605 µg/L, respectively.

CDFG (1992d) - In 1992, a 96-h static renewal toxicity test was performed by the CDFG Aquatic Toxicology Laboratory on technical diazinon (87%) with neonate mysid *Neomysis mercedis*. ASTM (1988a) standards were used. Five concentrations of diazinon were tested and solvent and water controls were used. Concentrations were measured during the test. Water quality parameters during the test averaged: temperature of 17.5 °C; pH of 8.36; hardness of 465.3 mg/L; salinity 1.73$^{o}/_{oo}$; conductivity 2932 µs/cm; dissolved oxygen of 8.92 mg/L. Control survival was 100%. The 96-h LC_{50} value was 4.82 µg/L. The NOEC and LOEC values, based on immobilization, were 2.45 µg/L and 4.5 µg/L, respectively.

Fernandez-Casalderry et al. (1992a) - In 1989, a 24-h static toxicity test was performed by the University of Valencia, Spain on technical grade diazinon (92%) with rotifer *Brachionus calyciflorus*. No commonly recognized test standards were used. Five concentrations were tested with nine replicates and a solvent control was included. Concentrations were not measured during the test and nominal concentrations were not given. Water quality parameters during testing were: temperature of 25^{o}C, pH of 7.4-7.8, and hardness of 80-100 mg/L. Control survival was 100%. The 24-h LC_{50} value was 29,220 µg/L.

Keizer et al. (1991) - In 1991, 96-h static toxicity tests were performed by the Instituto Superiore di Sanita Biochemical Toxicology Unit in Rome, Italy on technical grade diazinon (98%) with adult guppy *Poecilia reticulata* and adult zebrafish *Brachydanio rerio*. European Economic Community (1979) test methods were used. Seven concentrations were tested with three replicates for guppy and two replicates for zebrafish, and solvent controls were used.

Concentrations were measured daily but concentrations were not given. Water quality parameters during testing averaged: temperatures of 20-22 °C; pH of 7.6; dissolved oxygen of 6-9 mg/L. Control survival was 100%. The 96-h LC_{50} values were guppy: 800 µg/L and zebrafish: 8,000 µg/L.

Mayer and Ellersieck (1986), Dwyer and Sappington (pers. comm.) - From 1965 to 1985, 48-h and 96-h static toxicity tests were performed by the Columbia National Fisheries Laboratory of the U.S. Fish and Wildlife Service on technical grade diazinon (89-92%) with first instar cladoceran *Daphnia pulex*, mature amphipod *Gammarus fasciatus*, second year class stonefly *Pteronarcys californica*, bluegill *Lepomis macrochirus*, cutthroat trout *Oncorhynchus clarki* (two tests), lake trout *Salvelinus namaycush*, and rainbow trout *Oncorhynchus mykiss*. ASTM (1980) test standards were used. Four or more concentrations were tested in replicate and solvent (acetone) controls were used. Diazinon concentrations were not measured during the tests. Water quality parameters during the tests averaged: temperature of 21^{o}C (cladocerans), 21^{o}C (amphipod), 15^{o}C (stonefly), 18^{o}C (bluegill), 12^{o}C and 10^{o}C (cutthroat trout), 12^{o}C (lake trout) and 13^{o}C (rainbow trout); pH of 7.1-7.4; and hardness of 44-162 mg/L as $CaCO_3$. Control survival was acceptable in all tests. The 48-h EC_{50} value for cladoceran mortality and morbidity was 0.8 µg/L. The 96-h LC_{50} values were; bluegill: 168 µg/L, amphipod: 0.2 µg/L, stonefly: 25 µg/L, cutthroat trout: 2,760 µg/L and 1700 µg/L, lake trout: 602 µg/L, and rainbow trout: 90 µg/L. Although dissolved oxygen levels were not given, these tests were accepted because control survival was acceptable and ASTM standards were used.

Nimmo et al. (1981) - In 1980, a 96-h static toxicity test was performed by Environmental Research and Technology, Inc. and the EPA at Gulf Breeze, Florida on technical grade (percent active ingredient not given) diazinon on ≤48-h old mysids *Mysidopsis bahia*. EPA (1978) test standards were used. Five concentrations of diazinon were tested with four replicates and water and solvent controls. Concentrations were not measured during the test. Water quality parameters during the test averaged: temperature of 22-25 °C; pH of 8.0-8.2; and dissolved oxygen of 60% saturation. Control survival was 90%. The 96-h LC_{50} value was 4.82 µg/L.

Sanders and Cope (1966) - In 1966, 48-h static toxicity tests were performed by Fish-Pesticide Research Laboratory, Bureau of Sport Fisheries and Wildlife in Denver, Colorado on technical grade diazinon (percent active ingredient not given) with first instar larvae cladocerans *Daphnia pulex* and *Simocephalus serrulatus* (two tests). No commonly recognized test standard was mentioned. Four concentrations were used for *D. pulex* and five concentrations for *S. serrulatus*. Each test used a water control. Concentrations were not measured during the test. Water quality parameters during the tests averaged: temperatures of 60°C *Daphnia pulex*, 60°C and 70°C *Simocephalus serrulatus*; pH of 7.4-7.8; hardness and alkalinity not mentioned. Test water was aerated. Control survival was 100%. The 48-h EC_{50} values based on immobilization were: *Daphnia pulex*: 0.90 µg/L and *Simocephalus serrulatus:* 1.80 µg/L and 1.40 µg/L.

Surprenant (1988a) - In 1988, a 96-h flow-through toxicity test was performed by Springborn Life Sciences on technical grade diazinon (87.7%) with ≤24-h old mysids *Mysidopsis bahia*. Test methods used were similar to EPA (1985b) methods. Five concentrations of diazinon were tested in replicate and solvent and water controls were used. Concentrations were measured at the beginning and end of testing and measured concentrations averaged 78-100% of nominal concentrations. Water quality parameters during the test ranged: temperature of 25 ±1°C; pH of 7.8-7.9; dissolved oxygen of 5.6-7.3 mg/L; and salinity of 30-32°/oo. Control survival was 100%. The 96-h LC_{50} value for mysids was 4.2 µg/L.

Surprenant (1988b) - In 1988, a 96-h flow-through toxicity test was performed by Springborn Life Sciences on technical grade diazinon (87.7%) with eastern oyster *Crassostrea virginica*. Test methods used were similar to EPA (1985b) methods. Five concentrations of diazinon were tested in replicate and solvent and water controls were used. Concentrations were measured at the beginning and end of testing and measured concentrations averaged 55-88% of nominal concentrations. Water quality parameters during the test were: temperature of 20 ±2°C; pH of 7.4-8.1; dissolved oxygen of 5.8-7.7 mg/L; and salinity of 30-32°/oo. Control survival was 100%. The 96-h EC_{50} value (effect not given) for eastern oyster was 880 µg/L.

Vial (1990) - In 1990, 48-h static renewal toxicity tests were performed by Ciba-Geigy in Basel, Switzerland on technical diazinon (96%) with <24-h old cladoceran *Daphnia magna*. Organization for Economic Cooperation and Development (OECD) (1984) test standards were used. Six concentrations were tested with ten replicates and a water control. Concentrations were not measured during the test. Water quality parameters during the test were: temperature of 20 ±1°C; pH ranged from 7.8 to 9.3; dissolved oxygen ranged from 96 to 130% saturation; and hardness of 240 mg/L $CaCO_3$. Control survival was 100% The 48-h EC_{50} value for cladoceran *Daphnia magna* based on immobilization was >2.6 µg/L.

Unaccepted acute toxicity tests - The following tests for diazinon evaluated in Menconi and Cox (1994) did not use accepted test methods and/or produce accepted results.

Ebere and Akintonwa (1992) - In 1992, 96-hr static toxicity tests were performed by University of Lagos, Nigeria on technical grade diazinon (percent active ingredient not given) with fingerling *Gobius sp*, and juvenile *Desmocaris tirspimosa* and *Palaemontes africanus*. No commonly recognized test standards were mentioned. At least four concentrations of diazinon were tested and a water control was used. No mention of replicates was made. Concentrations were not measured during the test. Water quality parameters for the *Gobius sp*. and *Desmocaris tirspimosa* freshwater tests were: temperature of 25-27°C; dissolved oxygen of 8.1 ppm; conductivity of 90 µmho/cm; alkalinity of 20.4 mg/L. Water quality parameters for the brackish water averaged: dissolved oxygen of 4.3 ppm; salinity of 17 $^{o}/_{oo}$; conductivity of 38,000 µmhocm^{-1}; alkalinity of 60 mg/L. Control survival was >90%. The 96-h LC_{50} values were: *Gobius* sp.: 0.04 µl/L; *Palaemontes africanus*: 17.9 µl/L and *Desmocaris trispimosa*: 20.8 µl/L. These values were not accepted because concentrations were given in µl/L and it was not possible to convert to µg/L because percent active ingredient was not given. Attempts to obtain the necessary information from the researcher were not successful.

Federle and Collins (1975) - In 1975, a 96-h toxicity test was performed by Ohio State University on diazinon (percent active ingredient not given) with late instar damselfly *Lestes congener* nymphs. No commonly recognized test standards were mentioned and test dynamics were not given. Four concentrations were tested and solvent controls were used. Concentrations were not measured during the test. Water quality parameters during the tests were: temperature of 25 ±0.2°C; pH of 7.4. Dissolved oxygen levels, water hardness, and control survival were not given. The 96-h LC_{50} value was estimated to be 50 µg/L. This test was not accepted because the LC_{50} value was not calculated and essential information such as dissolved oxygen levels, water hardness, and control survival were not given.

Fernandez-Casalderrey et al. (1992b) - In 1992, a static toxicity test was performed on diazinon (92%) with 0-2 hr old rotifers *Brachionus calyciflorus pallas*. No commonly recognized test standards were mentioned and test duration was not given. Four concentrations were tested with four replicates per concentration. A solvent control was used. Concentrations were not measured during the test. Water quality parameters during the test were: temperature of 25°C; pH of 7.4-7.8; hardness of 80-100 mg/L; alkalinity of 60-70 mg/L as $CaCO_3$. Dissolved oxygen levels and control survival were not given. No LC_{50} values, NOEC values, or LOEC values were given. This test was not accepted because essential information such as test duration and dissolved oxygen levels were not given and no toxicity values were determined.

Ferrando et al. (1991) - In 1991, a 96-h flow-through toxicity test was performed by the University of Valencia, Spain on technical grade diazinon (92%) with European eel *Anguilla anguilla*. Life stage was not given. EPA (1975) standards were used. Four concentrations were tested and solvent controls were used. Concentrations were not measured during the test. Water quality parameters during the test were: temperature of 20°C; pH of 7.9±0.2; hardness of 250 mg/L; alkalinity of 4.1 mmol/L; dissolved oxygen not given. Control survival was 100%. The 96-h LC_{50} value was 80 µg/L. This test was not accepted because essential information such as dissolved oxygen levels, concentration scale, and mortality at each concentration was lacking. Efforts to obtain the missing information from the author were not successful.

Goodman et al. (1979) - In 1979, a 96-h flow-through toxicity test was performed by the EPA Environmental Research Laboratory in Gulf Breeze, Florida on diazinon (92.6%) with juvenile sheepshead minnow *Cyprinodon variegatus*. No commonly recognized test standards were mentioned. Five concentrations were tested and a solvent control was included. Concentrations were measured during the test. Water quality parameters during the test averaged: temperature of 30 ±2°C; pH not given; salinity of $22.7^{o}/_{oo}$. The test solutions were aerated and control survival was 100%. The 96-h LC_{50} value was 1470 µg/L. This was not used the concentrations tested did not produce an adequate mortality range.

Hashimoto et al. (1982) - In 1982, 24-h static toxicity tests were performed by Tokai Regional Fisheries Research Laboratory in Japan on diazinon (percent active ingredient not given) with eight life stages of carp *Cyprinus carpio*. No commonly recognized test standards were mentioned. Number of concentrations tested and use of controls were not mentioned. Concentrations were not measured during the test. Water quality parameters during the test were: temperature of 25 ±2°C; pH of 6.9-7.2. Dissolved oxygen, water hardness, and control survival were not given. The 24-h LC_{50} values were: eyed egg: 7.2 µg/L; sac fry: 6.1 µg/L; floating fry: 2.5 µg/L; one week old: 2.7 µg/L; two weeks old: 2.8 µg/L; four weeks old: 2.3 µg/L; eight weeks old; 1.9 µg/L and eleven weeks old: 2.4 µg/L. These tests were not accepted because essential information such as concentrations tested and use of controls was not given, and test duration was too short.

Khattat and Farley (1976) - In 1976, a 96-h static toxicity test was performed by the EPA Environmental Research Laboratory at Narragansett, Rhode Island on technical grade diazinon (97.6%) with adult marine copepod *Acartia tonsa*. No commonly recognized test standards were mentioned. Seven concentrations were tested with four replicates and solvent and water controls were used. Concentrations were measured at 24-h intervals. Water quality parameters during the test averaged: dissolved oxygen of 80% saturation; temperature of 17 ±1°C; salinity of $20^{o}/_{oo}$. Water and solvent control survival were 86.2% and 85.0%, respectively. The 96-h LC_{50} value was 2.57 µg/L. This test was unacceptable because control survival for both water and solvent was less than 90%.

Morgan (1976) - In 1975, a 168-h static toxicity test was performed by the University of Guelph on diazinon (50%) with midges *Chironomus tentans*. No test standards were mentioned. Three replicates were tested. Concentrations were measured. The temperature averaged 16^{o}C. Other water quality parameters were not measured. Control survival was not given. The 168-h LC_{50} for the midge was 0.027 µg/L. This value was not used because the pesticide formulation was too low in active ingredient and essential information, such as control survival and mortality range in the treatments, was not given.

Robertson and Mazella (1989) - In 1987, a 96-h static renewal toxicity test was performed by East Carolina University at Greenville, North Carolina on technical grade diazinon (88.6%) with freshwater snail *Gillia altilis*. Life stage was not given, and no commonly recognized test standards were mentioned. Four concentrations of diazinon were tested in triplicate and a solvent control was used. Concentrations were not measured during the test. Water quality parameters during the test were: temperature of 22 $\pm$1.5°C, pH of 6.7-6.9; dissolved oxygen of 8-11 mg/L; and hardness of 22-35 mg/L. Control survival was not given. The 96-h LC_{50} value was 11,000 µg/L. This test was not accepted because dechlorinated water was used and essential information such as control survival was not given.

Surprenant (1987a) - In 1987, a 96-h static toxicity test was performed on diazinon (48%) using bluegill *Lepomis macrochirus*. Life stage was not given. Test methods similar to ASTM (1980) methods were used. Five concentrations were tested in replicate with solvent controls. Concentrations were measured during the test. Water quality parameters during the test were: temperature of 22 $\pm$1 °C; pH of 7.6; dissolved oxygen of 62-103% saturation; hardness of 50 mg/L; conductivity of 90-140 umhos/cm; and alkalinity of 35 mg/L. Control survival was 100%. The 96-h LC_{50} value was 200 µg/L. This value was not used because the diazinon formulation was too low in active ingredient.

Surprenant (1987b) - In 1987, a 48-h static toxicity test was performed on diazinon (48%) with $\leq$24-hr old cladoceran *Daphnia magna*. Test methods similar to ASTM (1980) methods were used. Seven concentrations were tested with two replicates and solvent controls were used. Concentrations were measured twice during the test and averaged 82-100% of nominal concentrations. Water quality parameters during the test were: temperature of 21 $\pm$1°C; pH of 7.5-8.5; dissolved oxygen of 82-104% saturation; hardness of 180 mg/L; and alkalinity of 124 mg/L. Control survival was 100%. The 48-h LC_{50} value was 1.1 µg/L. This value was not used because the diazinon formulation was too low in active ingredient.

Surprenant (1987c) - In 1987, a 96-hr static toxicity test was performed on diazinon (48%) using rainbow trout *Oncorhynchus mykiss*. Life stage was not given. Test methods similar to ASTM (1980) methods were used. Seven concentrations were tested in replicate and solvent controls were used. Measured concentrations averaged 74-102% of nominal concentrations. Concentrations decreased 41% during the test. Water quality parameters during the test were: temperature of 13°C; pH of 7.1-8.1; dissolved oxygen of 66-98% saturation; and water hardness of 50 mg/L. Control survival was 100%. The 96-h LC_{50} value was 1,800 µg/L. This value was not used because the diazinon formulation was too low in active ingredient and the concentrations tested did not produce an adequate range of mortality.

Union Carbide (1978a) - In 1978, a 96-h static toxicity test was performed on diazinon (23%) using four month old bluegill *Lepomis macrochirus*. EPA (1975) test standards were used. Five concentrations were tested and a water control was included. Concentrations were not measured during the test. Water quality parameters during the test were: temperature of 21.8 ±0.3°C; pH of 7.46; dissolved oxygen of 4.4-9.0 mg/L; hardness of 44 mg/L; and conductivity of 32 mg/L. Control survival was not given. The 96-h LC_{50} value was 28,600 µg/L. The 96-h NOEC value (effects not given) was <18,000 µg/L. These values were not used because the formulation was too low in active ingredient, dissolved oxygen levels were too low, and control survival was not given.

Union Carbide (1978b) - In 1978, a 48-h static toxicity test was performed on diazinon (23%) using first instar cladoceran *Daphnia magna*. EPA (1975) test standards were used. Five concentrations were tested with four replicates per treatment. A water control was included. Concentrations were not measured; nominal concentrations ranged from 1.0-10 µg/L. Water quality parameters during the test were: temperature of 20°C; pH of 8.39; dissolved oxygen of 8.4-8.8 mg/L; and hardness of 252 mg/L. Control survival was 95%. The 48-h LC_{50} value was 5.03 µg/L. The NOEC value at 48-h was 3.2 µg/L. These values were not used because the diazinon formulation was too low in active ingredient.

Union Carbide (1978c) - In 1978, 96-h static toxicity test was performed on diazinon (23%) using four-month old rainbow trout *Oncorhynchus mykiss*. EPA (1975) test standards were used. Five concentrations were tested and a water control was included. Concentrations were not measured during the test. Water quality parameters during the test were: temperature of 11.8 ±0.3°C; pH of 7.47; dissolved oxygen of 4.1-9.0 mg/L; hardness of 44 mg/L; and alkalinity of 32 mg/L. Control survival was 100%. The 96-h LC_{50} value was 60,300 µg/L. The NOEC value at 96-h was <32,000 µg/L. These values were not used because the formulation was too low in active ingredient.

Vilkas (1976) - In 1986, a 48-h static toxicity test was performed on technical grade diazinon (percent active ingredient not given) with <20-h old cladocerans *Daphnia magna*. EPA (1975) and APHA (1971) test standards were used. Five concentrations were tested with four replicates and solvent controls were used. Concentrations were not measured during the test. Water quality parameters during the test were: temperature of 17°C; pH of 7.9; dissolved oxygen of 8.6-9.2 ppm; hardness of 50 mg/L; conductivity of 140 umhos/cm; and alkalinity of 25 mg/L. Control survival was 100%. The 48-h LC_{50} value was 0.98 µg/L. This test was not accepted because the concentrations tested did not produce an adequate mortality range.

Accepted chronic toxicity tests - The following tests for diazinon evaluated in Menconi and Cox (1994) used accepted test methods.

Nimmo et al. (1981), Nimmo (pers. comm.) - In 1980, a 28-d static toxicity test was performed by Environmental Research and Technology Inc. and the EPA in Gulf Breeze, Florida on technical grade diazinon (percent active ingredient not given) with ≤48-h old neonate mysid *Mysidopsis bahia*. EPA (1978) test standards were used. Five concentrations were tested with four replicates per treatment, and solvent and dilution water controls. Concentrations were measured weekly during the test. Water quality parameters during the tests were: temperature of 22-25°C; pH of 8.0-8.2; dissolved oxygen of 60% saturation. Control survival was >80%. The 28-d NOEC, LOEC, and MATC values based on reduced growth and fecundity were 1.15 µg/L, 3.27 µg/L, and 1.94 µg/L, respectively.

Surprenant (1988c) - In 1988, a 21-d flow-through toxicity test was performed by Springborn Life Sciences, Inc. on technical grade diazinon (87.7%) with ≤24-h old cladoceran *Daphnia magna*. ASTM (1980) and APHA (1985) test standards were used. Five concentrations were tested with four replicates per treatment and solvent and water controls were used. Concentrations were measured weekly during the test and measured concentrations averaged 64% of nominal concentrations. Water quality parameters during the test were: temperature of 20 ±1°C; pH of 7.9-8.3; dissolved oxygen of >60% saturation; hardness of 160-180 mg/L; and conductivity of 400-600 µmhos/cm. Control survival was >95% The 21-d NOEC, LOEC, and MATC values based on immobilization were 0.17 µg/L, 0.32 µg/L, and 0.23 µg/L, respectively.

Surprenant (1988d) - In 1988, a 34-d flow-through toxicity test was performed by Springborn Life Sciences Inc. on technical grade diazinon (87.7%) with embryo and larval fathead minnow *Pimephales promelas*. APHA (1985) and EPA (1981) test standards were used. Five concentrations were tested in replicate and water and solvent controls were used. Concentrations were measured and were 98-135% of nominal concentrations. Water quality parameters during the test were: temperature of 25 ±1°C; pH of 7.0-7.6; dissolved oxygen of 7.9-8.6 mg/L; hardness of 27-34 mg/L. Control survival was >90%. The 34-d NOEC, LOEC, and MATC values based on growth were 92 µg/L, 170 µg/L, and 125 µg/L, respectively.

Unaccepted chronic toxicity tests - The following tests for diazinon evaluated in Menconi and Cox (1994) did not use accepted test methods and/or produce acceptable results

Allison and Hermanutz (1977) - In 1977, a 274-d flow-through adult survival and reproduction test and a 60-d progeny (from tested adults) growth test were performed by the EPA Environmental Research Laboratory at Duluth, Minnesota on technical grade diazinon (92.5%) with fathead minnow *Pimephales promelas.* The EPA also performed a 173-d adult survival and reproduction test and a 122-d progeny test (from tested adults) on brook trout *Salvelinus fontinalis*. APHA (1971) test standards were used. Five concentrations were tested in replicate and a solvent control was used. Concentrations were measured weekly and averaged 73 to 110% of nominal concentrations. Water quality parameters during the tests were: temperature of 25 $\pm 1^{o}$C (minnows) and 12 $\pm 1^{o}$C (trout); pH of 7.3-7.5; dissolved oxygen of 85-86% saturation; and hardness of 44-45 mg/L. Control survival was 67% and 93% for progeny and adult fathead minnow, respectively. Control survival was 89% and 100% for progeny and adult brook trout, respectively. The lowest concentration tested on fathead minnow, 3.2 µg/L, had a significant detrimental effect on hatching success and caused scoliosis in progeny. The lowest concentration tested on brook trout, 0.8 µg/L, had a significant detrimental effect on progeny growth. These values were not used because no NOEC value was determined. One fathead minnow test was terminated at 91-d because the concentrations were too high. Acute tests were also conducted with fathead minnow, bluegill, brook trout, and flagfish.

Bresch (1991), Bresch (pers. comm.) - In 1991, 28-d and 42-d flow-through toxicity tests were performed by the Institute for Hygiene and Toxicology, Germany on analytical grade diazinon (percent active ingredient not given) with adult zebrafish *Brachydanio rerio* and fingerling rainbow trout *Oncorhynchus mykiss*. EPA (1982), ASTM (1988b), and OECD (1983) test standards were used. Three concentrations of diazinon were tested in replicate and a water control was used. Concentrations were measured weekly and measured concentrations averaged 20% of nominal concentrations. Water quality parameters during the tests were: temperatures of 24-26°C (zebrafish) and 15-17°C (rainbow trout); pH of 7.4; dissolved oxygen of >60% saturation (zebrafish); >70% saturation (rainbow trout); and hardness of 360 mg/L. Control survival was 86.5% (zebrafish); 100% (rainbow trout). The zebrafish NOEC and LOEC values, based on growth (length) reduction, were 750 µg/L and 1,500 µg/L, respectively. No rainbow trout died during the test, however abnormal fish were present in all groups. The zebrafish test was not accepted because the test did not have a sufficient number of concentrations. The rainbow trout test was not accepted because it did not have a sufficient number of concentrations, no pertinent values were obtained, and an insufficient number of trout were tested.

Goodman et al. (1979) - In 1979, a 108-d flow-through toxicity test was performed by the EPA Environmental Research Laboratory in Gulf Breeze, Florida on diazinon (92.6%) using juvenile sheepshead minnow *Cyprinodon variegatus*. No commonly recognized testing standards were mentioned. Five concentrations and a solvent control were tested. Measured concentrations were 65-78% of nominal concentrations. Water quality parameters during the test were: temperature of 30 $\pm 2^{o}$C; pH not given; salinity averaged $16.5^{o}/_{oo}$; test solutions were aerated; water hardness not given. Control survival was not given. Fecundity decreased at the lowest concentration tested (0.47 µg/L) and darkened flesh appeared at concentrations between 3.5 µg/L and 6.5 µg/L. These values were not used because no NOEC value was determined.

Morgan (1976) - In 1975, a 113-d static toxicity test was performed by the University of Guelph on diazinon (50 %) with midges *Chironomus tentans*. No test standards were mentioned. Six replicates were tested. Concentrations were measured. The temperature ranged from 22 to 25^{o}C. Other water quality parameters were not measured. Control survival was not given. The effects were not given. This test was not accepted because the pesticide formulation was too low in active ingredient; essential information, such as control survival and mortality range, was not given, and effects were not determined.

Vial (1990) - In 1990, 22-d static renewal toxicity tests were performed by Ciba-Geigy in Basel, Switzerland on technical diazinon with <24-h old cladoceran *Daphnia magna*. OECD test standards were used. Six concentrations were tested with ten replicates and a water control. Concentrations were measured weekly during the test and measured concentrations ranged from 75 to 4215% of nominal concentrations. Water quality parameters during the test were: temperature of 20 $\pm 1^{o}$ C; pH ranged from 7.8 to 9.3; dissolved oxygen ranged from 96 to 130% saturation; and hardness of 240 mg/L $CaCO_3$. Control survival was 100%. The EC_{50} value for cladoceran *Daphnia magna*, based on immobilization, was >2.6 µg/L. The LOEC value, based on number of young produced, was 0.0026 µg/L. The NOEC value was reported as <0.0026 µg/L, and no MATC could be calculated. The chronic toxicity data produced by this study could not be used because no definite NOEC or MATC values were determined. The LOEC value was 100-fold lower than that obtained by Surprenant (1988c). In addition, the nominal and measured concentrations differed too much to be accepted for a chronic study. However, the acute toxicity data were accepted.

APPENDIX D. Abstracts of acute toxicity tests for chlorpyrifos evaluated in Menconi and Paul (1994).

Accepted acute toxicity tests - The following tests for chlorpyrifos evaluated in Menconi and Paul (1994) used accepted test methods.

Borthwick and Walsh (1981) - In 1981, 96-h static toxicity tests were performed by the U.S. EPA on technical grade Dursban^R (97.7%) with juvenile mysid *Mysidopsis bahia* and 28-d old fry sheepshead minnow *Cyprinodon variegatus*. As part of this series of tests, 48-h static toxicity tests were performed with <2-h old eastern oyster *Crassostrea virginica*. ASTM (1978) testing guidelines were followed. Five concentrations of Dursban^R and solvent and dilution water controls were tested with mysids and sheepshead minnows. Seven concentrations of Dursban^R were tested with eastern oysters. Two replicates per concentration were tested with mysids and sheepshead minnows. Four replicates per concentration were tested with eastern oysters. Measurement of chlorpyrifos concentrations was not mentioned for any test. Water quality parameters during the eastern oyster, mysid, and sheepshead minnow tests averaged: temperature of 25.0 $\pm$1^{o}C; Ph was not mentioned; dissolved oxygen level was not measured; and salinity of 20$^o/_{oo}$. Control survival was greater than 90% for all tests. The 96-h LC_{50} values for the mysid and the sheepshead minnow were 0.056 µg/L and 270 µg/L, respectively. The 96-h EC_{50} value, based on abnormal development, for the eastern oyster was 1991 µg/L. Tests were also conducted using diatoms *Skeletonema costatum, Isochrysis galbana*, and *Thalassiosira pseudonana*.

Borthwick et al. (1985) - In 1985, 96-h flow-through toxicity tests were performed by the U.S. EPA on technical grade chlorpyrifos (92%) with 0-d, 7-d, 14-d, and 28-d old California grunion *Leuresthes tenuis*, Atlantic silverside *Menidia menidia*, and tidewater silverside *Menidia peninsulae*. ASTM (1980) testing guidelines were followed. Five concentrations of chlorpyrifos were tested and solvent and dilution water controls were used. The number of replicates tested was not mentioned. Concentrations were measured. Water quality parameters during the test averaged: temperature of 25^{o}C; pH was not mentioned; dissolved oxygen level was not mentioned; and salinity of 20$^o/_{oo}$ in the *M. menidia* and *M. peninsulae* tests and 25$^o/_{oo}$ in the *L. tenuis* tests. Control survival was not mentioned. The 96-h LC_{50} values for the 0-d, 7-d, 14-d, and 28-d old grunion were 1.0 µg/L, 1.0 µg/L, 1.0 µg/L and 1.3 µg/L, respectively. The 96-h LC_{50} values for the 0-d, 7-d, 14-d and 28-d old Atlantic silverside were 0.5 µg/L, 1.0 µg/L, 1.1 µg/L, and 3.0 µg/L, respectively. The 96-h LC_{50} values for the 0-d, 7-d, 14-d and 28-d old tidewater silversides were 1.0 µg/L, 0.5 µg/L, 0.4 µg/L, 0.9 µg/L, respectively.

California Department of Fish and Game (CDFG) (1992e) - In 1992, 96-h static renewal toxicity tests (Test No. 92-133) were performed on technical grade chlorpyrifos (99%) with neonate mysid *Neomysis mercedis*. ASTM (1992) testing guidelines were followed. Five concentrations of chlorpyrifos were tested and solvent and dilution water controls were used. Two replicates per concentration were tested. Chlorpyrifos concentrations were measured at the beginning and end of each test and averaged 86 to 124% of nominal concentrations. Water quality parameters during the test averaged: temperature of 17.2°C; pH of 8.4; dissolved oxygen level of 8.4 mg/L; hardness of 499.0 mg/L; conductivity of 3096 µs/cm; and alkalinity of 154.0 mg/L. Control survival was 95% in the solvent control and 100% in the dilution water control. The 96-h LC_{50} value for *N. mercedis* was 0.16 µg/L.

CDFG (1992e) - In 1992, 96-h static renewal toxicity tests (Test No. 92-142) were performed on technical grade chlorpyrifos (99%) with neonate mysid *Neomysis mercedis*. ASTM (1992) testing guidelines were followed. Five concentrations of chlorpyrifos were tested and solvent and dilution water controls were used. Two replicates per concentration were tested. Chlorpyrifos concentrations were measured at the beginning and end of each test and averaged 71 to 84% of nominal concentrations. Water quality parameters during the test averaged: temperature of 17.1°C; pH of 8.4; dissolved oxygen level of 9.3 mg/L; hardness of 509.0 mg/L; conductivity of 3151 µs/cm; and alkalinity of 151.0 mg/L. Control survival was 100% for both solvent and dilution water controls. The 96-h LC_{50} value for *N. mercedis* was 0.14 µg/L.

CDFG (1992e) - In 1992, 96-h static renewal toxicity tests (Test No. 92-143) were performed by CDFG on technical grade chlorpyrifos (99%) with neonate mysid *Neomysis mercedis*. ASTM (1992) testing guidelines were followed. Five concentrations of chlorpyrifos were tested and solvent and dilution water controls were used. Two replicates per concentration were tested. Chlorpyrifos concentrations were measured at the beginning and end of each test and averaged 71 to 84% of nominal concentrations. Water quality parameters during the test averaged: temperature of 17.4°C; pH of 8.2; dissolved oxygen level of 8.9 mg/L; hardness of 515.0 mg/L; conductivity of 3192 µs/cm; and alkalinity of 151.5 mg/L. Control survival was 100% for both solvent and dilution water controls. The 96-h LC_{50} value for *N. mercedis* was 0.15 µg/L.

CDFG (1992f) - In 1992, 96-h static toxicity tests (Test No. 92-139) were performed by the CDFG on technical grade chlorpyrifos (99%) with neonate cladoceran *Ceriodaphnia dubia*. EPA (1989) and ASTM (1988a,b) testing guidelines were followed. Five concentrations of chlorpyrifos were tested and solvent and dilution water controls were used. Nine replicates per concentration were tested. Chlorpyrifos concentrations were measured at the beginning and end of each test and averaged 75 to 100% of nominal concentrations. Water quality parameters during the test averaged: temperature of 24.3°C, pH of 8.2; dissolved oxygen level of 7.7 mg/L; hardness of 121.5 mg/L; conductivity of 333.7 µs/cm; and alkalinity of 105.0 mg/L. Control survival was 90% in both the solvent and dilution water controls. The 96-h LC_{50} value for *C. dubia* was 0.08 µg/L.

CDFG (1992f) - In 1992, 96-h static toxicity tests (Test No. 92-150) were performed by the CDFG on technical grade chlorpyrifos (99%) with neonate cladoceran *Ceriodaphnia dubia*. EPA (1989) and ASTM (1988a,b) testing guidelines were followed. Five concentrations of chlorpyrifos were tested and solvent and dilution water controls were used. Nine replicates per concentration were tested. Chlorpyrifos concentrations were measured at the beginning and end of each test and averaged 94 to 105% of nominal concentrations. Water quality parameters during the test averaged: temperature of 24.6^{o}C; pH of 8.3; dissolved oxygen level of 7.7 mg/L; hardness of 120.0 mg/L; conductivity of 325.7 µs/cm; and alkalinity of 107.0 mg/L. Control survival was 100% in both solvent and dilution water controls. The 96-h LC_{50} value for *C. dubia* was 0.13 µg/L.

Clark et al. (1985) - In 1985, 96-h static and flow-through toxicity tests were performed by the U.S. EPA on technical grade chlorpyrifos (percent active ingredient not specified) with tidewater silverside *Menidia peninsulae* and inland silverside *Menidia beryllina*. ASTM (1980) testing guidelines were followed. Five concentrations of chlorpyrifos were tested and solvent and dilution water controls were used. One replicate per concentration was tested. Chlorpyrifos concentrations were measured at 48-h and 96-h but values were not mentioned. Water temperatures in the tidewater and inland silverside tests averaged 24.6^{o}C and 24.5^{o}C, respectively. Other water quality parameters during the test were measured but values were not given. Solvent and dilution water controls were used and survival was within ASTM guidelines. The 96-h LC_{50} values for tidewater silverside and inland silverside were 1.3 µg/L and 4.2 µg/L, respectively. Although this study had some deficiencies it was considered acceptable because of the reputation of the laboratory, ASTM guidelines were followed, and control survival was acceptable.

Federle and Collins (1976) - In 1976, 96-h static toxicity tests were performed by the Department of Entomology, Ohio State University on technical grade chlorpyrifos (94%) with adult crawling water beetle *Petodytes* sp. Commonly recognized testing guidelines were not mentioned. Four concentrations of chlorpyrifos were tested and a solvent control was used. Three replicates per concentration were tested. All concentrations tested were nominal. Water quality parameters during the test averaged: temperature of 25 $\pm$2^{o}C; pH of 7.4; dissolved oxygen level was not mentioned but test solutions were aerated. Control survival was 95%. The 96-h LC_{50} value for *Petodytes* sp. was 0.8 µg/L.

Hansen et al. (1986) - In 1986, 96-h static toxicity tests were performed by the U.S. Environmental Protection Agency on technical grade chlorpyrifos (92%) with 2-mo. old gulf toadfish *Opsanus beta*. ASTM (1985) proposed testing guidelines were followed. Five concentrations of chlorpyrifos were tested and solvent and dilution water controls were used. One replicate per concentration was tested. Chlorpyrifos concentrations averaged 50 to 60% of nominal concentrations. Water quality parameters during the test averaged: temperature of 25 to 27^{o}C; pH was not mentioned; dissolved oxygen was not measured; and salinity of 29$^{o}/_{oo}$ to 30$^{o}/_{oo}$. Control survival was 100%. The 96-h LC_{50} value for *Opsanus beta* was 520 µg/L.

Holcombe et al. (1982) - In 1982, a 96-h flow-through toxicity test was performed by the U.S. EPA on technical grade Dursban^R (99.9%) with juvenile rainbow trout *Oncorhynchus mykiss* and 31-d to 32-d old fathead minnow *Pimephales promelas*. APHA (1975) and EPA (1975) testing guidelines were followed. Five concentrations of Dursban^R and a water control were tested in each test. Two replicates per concentration were tested. Chlorpyrifos concentrations were measured daily and averaged 88 to 112% of nominal concentrations in rainbow trout tests and 84 to 116% in fathead minnow tests. Water quality parameters during the test averaged: temperature of 15.6 $\pm$1.8^{o}C for rainbow trout tests and 25.1 $\pm$1.3^{o}C for fathead minnow tests; pH of 7.0 to 7.4; dissolved oxygen level of 9.3 mg/L for rainbow trout tests and 7.3 mg/L for fathead minnow tests; hardness of 45.3 mg/L; and alkalinity of 41.8 mg/L. Control survival was 100% in both tests. The 96-h LC_{50} value for the fathead minnow was 203 µg/L. The 96-h LC_{50} value for the rainbow trout was 8.0 µg/L.

Jarvinen and Tanner (1982) - In 1982, 96-h flow-through toxicity tests were performed by the U.S. EPA on technical grade chlorpyrifos (98.7%) with larval fathead minnow *Pimephales promelas*. APHA (1975) testing guidelines were followed. Five concentrations of chlorpyrifos were tested and a dilution water control was used. Two replicates per concentration were tested. Measured concentrations averaged 102 to 144% of nominal concentrations. Water quality parameters during the test averaged: temperature of 25.0 $\pm$0.6^{o}C; pH of 7.4 to 7.8; dissolved oxygen level of 6.5 to 8.4 mg/L; hardness of 45.8 mg/L; and alkalinity of 43.1 mg/L. Control survival was 100%. The 96-h LC_{50} value for *P. promelas* was 140 µg/L.

Kersting and Van Wijngaarden (1992) - In 1992, a 48-h static toxicity test was performed by the Research Institute for Nature Management in The Netherlands on technical grade chlorpyrifos (99%) with <24-h old cladoceran *Daphnia magna*. Commonly recognized test guidelines were not mentioned. Six concentrations of chlorpyrifos were tested and solvent and dilution water controls were used. Two replicates per concentration were tested. Chlorpyrifos concentrations were measured at the beginning of each test and averaged 57% of nominal concentrations. Water quality parameters during the test averaged: temperature of 19.5 $\pm$0.5^{o}C; pH of 6.8 to 7.0; and dissolved oxygen level of 7.7 mg/L to 8.8 mg/L. Control survival was 100%. The 48-h LC_{50} value for *D. magna* was 1.0 µg/L.

Macek et al. (1969) - In 1969, 96-h static toxicity tests were performed by the U.S. Bureau of Sport Fisheries and Wildlife on technical grade Dursban^R (97%) with rainbow trout *Oncorhynchus mykiss*. Commonly recognized testing guidelines were not mentioned. Seven concentrations of chlorpyrifos and a solvent control were tested. One replicate per concentration was tested. All concentrations were nominal. Water quality parameters during the test averaged: temperature of 1.6 $\pm$0.6^{o}C in one series of tests, 7.2 $\pm$0.6^{o}C in a second series of tests, and 12.7 $\pm$0.6^{o}C in a third series of tests; pH of 7.1; dissolved oxygen level was not measured but solutions used in tests were well aerated; and alkalinity of 35 mg/L. Control survival was 100%. The 96-h LC_{50} value for *O. mykiss* at 1.6^{o}C was 51 µg/L. The 96-h LC_{50} value for *O. mykiss* at

7.2^{o}C was 15 µg/L. The 96-h LC_{50} values for *O. mykiss* at 12.7^{o}C was 7.1 µg/L. Only the test performed at 12.7 $\pm$0.6^o C was used because it most closely adhered to ASTM (1988a) standards for trout tests.

Mayer (1987) - In 1987, results of acute toxicity tests on 197 chemicals with 52 estuarine and marine species were compiled. All tests were performed at the Environmental Research Laboratory, Gulf Breeze, Florida during 1961 to 1986. The tests used technical grade chlorpyrifos (92%) and generally complied with ASTM (1980) standards. At least four concentrations of chlorpyrifos were tested in each test. Depending on the species, temperatures ranged from 11^{o}C to 31^{o}C. Dissolved oxygen, pH, control survival, water hardness, and chlorpyrifos concentrations were not given. The 96-h and 48-h LC_{50} values and the 48-h EC_{50} values are listed in Table A-1. Although information about some important test characteristics could not be obtained, most of these data were accepted because of the use of ASTM guidelines and the reputation of the laboratory. Acceptable data were available for Atlantic and tidewater silversides, blue crab, brown, grass, and pink shrimps, California grunion, gulf and longnose killifish, gulf toadfish, *Mysidopsis bahia*, sheepshead minnow, and striped mullet.

Mayer and Ellersieck (1986) - In 1986, a study was conducted by the Fish and Wildlife Service to generate static acute toxicity test data for 410 chemicals with 66 freshwater species. All tests were performed at the Columbia National Fisheries Research Laboratory and its field laboratories between 1965 to 1984. The studies on technical grade chlorpyrifos (97%) were conducted with eight species. The tests were generally in compliance with ASTM (1980) and EPA (1975) standards. At least five concentrations of chlorpyrifos were tested. Two replicates per concentration were tested. Depending on the species, water quality parameters during the tests were as follows: temperature of 2.0^{o}C to 29^{o}C; pH of 6.0 to 9.0; and hardness of 44 mg/L to 272 mg/L. Control survival, dissolved oxygen, and measurement of chlorpyrifos concentrations were not discussed. The 96-h LC_{50} values are listed in Table A-1. Although information about some important test characteristics could not be obtained, most of these data were accepted because of the use of ASTM guidelines and the reputation of the laboratory. Acceptable data were available for bluegill, channel catfish, and lake and cutthroat trouts.

Phipps and Holcombe (1985) - In 1985, 96-h flow-through toxicity tests were performed by the U.S. EPA on technical grade chlorpyrifos (99.9%) with rainbow trout *Oncorhynchus mykiss*, fathead minnow *Pimephales promelas*, goldfish *Carassius auratus*, channel catfish *Ictalurus punctatus*, bluegill *Lepomis macrochirus*, crayfish *Orconectes immunis*, and snail *Aplexa hypnorum*. ASTM (1980) and APHA (1980) testing guidelines were followed. Three or five concentrations of chlorpyrifos were tested and water controls were used. One replicate per concentration was tested in all tests. Chlorpyrifos concentrations were measured daily and averaged 85 to 114% of nominal concentrations. Water quality parameters during the test averaged: temperature of 17.3 $\pm$0.6^{o}C; pH of 7.1 to 7.8; dissolved oxygen level of 7.5 $\pm$1.6 mg/L; hardness of 44.4 mg/L; and alkalinity of 45.4 mg/L. Control survival was 100%. The 96-h LC_{50}

value for rainbow trout, fathead minnow, goldfish, channel catfish, bluegill, crayfish, and snails were 9 µg/L, 542 µg/L, >806 µg/L, 806 µg/L, 10 µg/L, 6.0 µg/L, and >806 µg/L, respectively.

Sanders (1969) - In 1969, 96-h static toxicity tests were performed by the U.S. Bureau of Sport Fisheries and Wildlife on technical grade Dursban^R (97%) with 2 month old (±5 days) amphipod *Gammarus lacustris*. Commonly recognized testing guidelines were not mentioned. Five concentrations of Dursban^R were tested and a dilution water control was used. One replicate per concentration was tested. Measurement of chlorpyrifos concentrations was not mentioned. Water quality parameters during the test averaged: temperature of 70 ±1^{o}F; pH of 7.1; dissolved oxygen was not measured but test water was aerated for 10 minutes before testing began; and alkalinity of 30.0 mg/L. Control survival was 100%. The 96-h LC_{50} value for *G. lacustris* was 0.11 µg/L.

Sanders and Cope (1968) - In 1968, 96-h static toxicity tests were performed by the U.S. Bureau of Sport Fisheries and Wildlife on technical grade Dursban^R (97%) with naiad stoneflies *Pteronarcys californica*, *Pteronarcella badia*, and *Claassenia sabulosa*. Commonly recognized testing guidelines were not mentioned. Four concentrations of Dursban^R were tested and a dilution water control was used. One replicate per concentration was tested. Measurement of chlorpyrifos concentrations was not mentioned. Water quality parameters during the test averaged: temperature of 15.5 ±0.5^{o}C; pH of 7.1; dissolved oxygen level of 7 mg/L initially, 5 mg/L after 24-h, and 3 mg/L at 96-h; alkalinity of 35 mg/L. Control survival was 100%. The 96-h LC_{50} values for *Pteronarcys californica*, *Pteronarcella badia*, and *Claassenia sabulosa* were 10 µg/L, 0.38 µg/L, and 0.57 µg/L, respectively. Although the dissolved oxygen was too low, these tests were accepted because control survival was 100%

Unaccepted acute toxicity tests - The following tests for chlorpyrifos evaluated in Menconi and Paul (1994) did not use accepted test methods and/or produce accepted results.

Acevedo (1991) - In 1991, 96-h static and flow-through toxicity tests were performed by the Hawaii Institute of Marine Biology on technical grade chlorpyrifos (percent active ingredient not specified) with coral planulae *Pocillopora damicornis*. EPA (1988) testing guidelines were followed. Four concentrations of chlorpyrifos were tested. Three replicates per concentration were tested. The use of controls was not mentioned. Measurement of chlorpyrifos concentrations was not mentioned. Water quality parameters during the study averaged: temperature of 25-27^oC; pH was not mentioned; dissolved oxygen level was not mentioned; and salinity was not mentioned. Control survival was not mentioned. The static and flow-through LC_{50} values were not reported. This test was not used because essential information, such as toxicity values, and control survival, was lacking.

Ali and Majori (1982) - In 1982, 24-h toxicity tests were performed by the University of Florida on technical grade chlorpyrifos (percent active ingredient not specified) with fourth instar larvae of midge *Chironomus salinarius*. Commonly recognized testing guidelines were not mentioned. Four to five concentrations and a dilution water control were tested. Three replicates per concentration were tested. Measurement of chlorpyrifos concentrations was not mentioned. Water quality parameters during the test averaged: temperature of 27 $\pm 2^oC$; pH of 7.8 to 9.0; dissolved oxygen level of <2 to 10 mg/L; and salinity of 27 to $45^o/_{oo}$. Control survival was not mentioned. The 24-h LC_{50} value for *C. salinarius* was 0.44 µg/L. This value was not used because the organisms had been exposed to pesticides prior to testing, dissolved oxygen levels fell below an acceptable range, and essential information, such as control survival, was lacking.

CDFG (1992f) - In 1992, 96-h static toxicity tests (Test No. 92-137) were performed by CDFG on technical grade chlorpyrifos (99%) with neonate cladoceran *Ceriodaphnia dubia*. EPA (1989) and ASTM (1992a,b) testing guidelines were followed. Five concentrations of chlorpyrifos were tested and solvent and dilution water controls were used. Nine replicates per concentration were tested. Chlorpyrifos concentrations were measured at the beginning and end of each test and averaged 105 to 150% of nominal concentrations. Water quality parameters during the test averaged: temperature of 24.5^oC; pH of 8.4; dissolved oxygen level of 7.7 mg/L; hardness of 119.0 mg/L; conductivity of 345.6 µs/cm; and alkalinity of 105.5 mg/L. Control survival was 70% in the solvent control and 100% in the dilution water control. The 96-h LC_{50} value for *C. dubia* was 0.12 µg/L. This value was not used because solvent control survival was less than 90%.

Carter and Graves (1973) - In 1973, 96-h static toxicity tests were performed by the Department of Entomology, Louisiana State University on chlorpyrifos (percent active ingredient not specified) with White River crayfish *Procambarus acutus*, bluegill *Lepomis macrochirus*, mosquitofish *Gambusia affinis*, and channel catfish *Ictalurus punctatus*. APHA (n.d.) testing guidelines were followed. The number of chlorpyrifos concentrations tested and use of controls were not mentioned. Five replicates per concentration were used in the crayfish tests. Two replicates per concentration were used in all other tests. Measurement of chlorpyrifos concentrations was not mentioned. Water quality parameters during the test averaged: temperature of 26°C in crayfish and channel catfish tests, 23°C in bluegill tests, and 24°C in mosquitofish tests; pH was not mentioned for any test; and dissolved oxygen level of 7 mg/L to 10 mg/L in bluegill and channel catfish tests, 9 mg/L to 11 mg/L in White River crawfish tests, and 9 mg/L in mosquitofish tests. The 96-h LC_{50} values for crayfish, bluegill, mosquitofish, and channel catfish were 2 µg/L, 30 µg/L, 280 µg/L, and 160 µg/L, respectively. These values were not used because essential information, such as control survival and concentrations tested, was lacking.

Cebrian et al. (1992) - In 1992, 96-h static toxicity tests were performed by the Department of Animal Biology, University of Valencia, Spain on technical grade chlorpyrifos (99.8%) with crayfish *Procambarus clarkii*. EPA (1975) testing guidelines were followed. The chlorpyrifos concentrations and number of replicates tested were not mentioned. A solvent control was used. Measurement of chlorpyrifos concentrations was not mentioned. Water quality parameters during the test averaged: temperature of 22 ±1°C; pH of 7.9 ±0.2; dissolved oxygen level was not mentioned; hardness of 250 mg/L as $CaCO_3$; and alkalinity of 4.1 mM/L. Control survival was not mentioned. The 96-h LC_{50} value for *P. clarkii* was 21 µg/L, respectively. This value was not used because essential information, such as control survival and concentrations tested, was lacking.

Darwazeh and Mulla (1974) - In 1974, 92-h toxicity tests were performed by the Department of Entomology, University of California at Riverside on technical chlorpyrifos (percent active ingredient not specified) with mosquitofish *Gambusia affinis*. Commonly recognized testing guidelines were not mentioned. The chlorpyrifos concentrations tested were not mentioned. Two replicates per concentration were tested. Measurement of chlorpyrifos concentrations was not mentioned. Water quality parameters during the test were not mentioned. Control survival was not mentioned. LC_{50} values were not determined in this study. The 92-h LC_{85} value for *G. affinis* was 1000 µg/L. The test was unacceptable because essential information, such as water quality parameters and concentrations tested, was lacking, the test duration was less than 96-h, and LC_{50} values were not determined.

Davey et al. (1976) - In 1976, 72-h static toxicity tests were performed by the Department of Entomology, University of Arkansas on chlorpyrifos (percent active ingredient not specified) with mature mosquitofish *Gambusia affinis* and green sunfish *Lepomis cyanellus*. Commonly recognized testing guidelines were not mentioned. One concentration of chlorpyrifos was tested and a solvent control was used. Two replicates per concentration were tested. Measurement of chlorpyrifos concentrations was not mentioned. Water quality parameters during the test were not mentioned. Control survival was not mentioned. The 72-h LC_{50} values for the mosquitofish and the green sunfish were 260 µg/L and 40 µg/L, respectively. These values were not used because the test duration was too short, an inadequate number of concentrations was tested, and essential information, such as water quality parameters and control survival, was lacking.

Earnest (1970) - In 1970, 96-h static and flow-through toxicity tests were performed by the U.S. Bureau of Sport Fisheries and Wildlife on technical grade Dursban^R (99%) with Korean shrimp *Palaemon macrodactylus*. A 96-h flow-through toxicity test was performed on Dursban^R (90%) with striped bass *Morone saxatilis*. Commonly recognized testing guidelines were not mentioned. Dursban^R concentrations, number of replicates, and use of controls were not mentioned. Measurement of chlorpyrifos concentrations was not mentioned. Water quality parameters during the test averaged: temperature of 12.2^{o}C to 12.8^{o}C; pH was not mentioned; dissolved oxygen level was not mentioned; and salinity of 15$^o/_{oo}$ to 30$^o/_{oo}$. Control survival was not mentioned. The 96-h static and flow-through TL_{50}s for the Korean shrimp were 0.25 µg/L and 0.01 µg/L, respectively. The 96-h flow-through TL_{50} for the striped bass was 0.58 µg/L. These values were not used because essential information, such as control survival and use of controls, was lacking.

El-Refai et al. (1976) - In 1976, 48-h static toxicity tests were performed by Al-Azhar University Cairo on Dursban^R (40.8%) with fingerling tilapia *Tilapia nilotica* and carp *Cyprinus carpio*. Commonly recognized testing guidelines were not mentioned. One replicate per concentration was tested when larger fish were used, otherwise replicates were not used. Chlorpyrifos concentrations were measured but values were not given. Water quality parameters during the test averaged: temperature of 22^{o}C to 25^{o}C; pH of 7.8 to 8.2; dissolved oxygen level of 6.8 mg/L to 7.4 mg/L; hardness of 116 mg/L to 123 mg/L; conductivity of 270 to 300 µmho/cm; and alkalinity of 122 mg/L to 125 mg/L. Control survival was not mentioned. For smaller carp, the 48-h LC_{50} value was 280 µg/L. For larger carp, the 48-h LC_{50} value was 59 µg/L. For smaller tilapia, the 48-h LC_{50} value was 62 µg/L. For larger tilapia, the 48-h LC_{50} value was 114 µg/L. These values were not used because essential information, such as control survival, was lacking, the test duration was less than 96-h, the pesticide formulation used was too low in active ingredient, and the temperature varied by more than 2^{o}C.

Ferguson et al. (1966) - In 1966, 36-h toxicity tests were performed by the Department of Zoology, Mississippi State University on technical grade chlorpyrifos (99 ±1%) with the golden shiner *Notemigonus crysoleucas*, mosquitofish *Gambusia affinis*, and green sunfish *Lepomis cyanellus*. Commonly recognized testing guidelines were not mentioned. Chlorpyrifos concentrations and number of replicates tested were not mentioned. Measurement of chlorpyrifos concentrations was not mentioned. Water quality parameters during the test averaged: temperature was not mentioned; pH of 7.4; dissolved oxygen level was not mentioned; and hardness of 24 mg/L. Control survival was 95%. The 36-h LC_{50} values for the golden shiner, mosquitofish, and green sunfish were 35 µg/L to 125 µg/L, 215 µg/L to 595 µg/L, and

22.5 µg/L to 125 µg/L, respectively. These values were not used because test duration was less than 96-h and organisms were exposed to pesticides prior to testing.

Ferrando et al. (1991) - In 1991, 96-h flow-through toxicity tests were performed by the Department of Animal Biology, University of Valencia, Spain on technical grade chlorpyrifos (97%) with the European eel *Anguilla anguilla*. U.S. EPA (1975) testing guidelines were followed. Chlorpyrifos concentrations tested were not mentioned. Solvent and dilution water controls were used. Three replicates per concentration were tested. Measurement of chlorpyrifos concentrations was not mentioned. Water quality parameters during the test averaged: temperature of 20^{o}C; pH of 7.9 $\pm$0.2; dissolved oxygen level was not mentioned; alkalinity of 4.1 mmol/L; and a hardness of 250 mg/L. Control survival was 100%. The 96-h LC_{50} value for *A. anguilla* was 540 µg/L. This value was not used because dissolved oxygen was not measured and essential information, such as concentrations tested, was lacking.

Ferrando and Andreu-Moliner (1991) - In 1991, 24-h static toxicity tests were performed by the Department of Animal Biology, University of Valencia, Spain on chlorpyrifos (percent active ingredient not specified) with newly hatched rotifera: *Brachionus calyciflorus* and *Brachionus plicatilis*. EPA (1985) testing guidelines were followed. Five concentrations of chlorpyrifos were tested. Use of a control was not mentioned. Three replicates per concentration were tested. Measurement of chlorpyrifos concentrations was not mentioned. Water quality parameters during the test averaged: temperature of 25^{o}C; pH of 7.4 to 7.8 in *B. calyciflorus* tests and 7.7 in *B. plicatilis* tests; hardness of 80 mg/L to 100 mg/L in *B. calyciflorus* tests (hardness not mentioned in *B. plicatilis* tests); and salinity of 15$^{o}/_{oo}$ in *B. plicatilis* tests (salinity not mentioned in B. calyciflorus tests). Control survival was 100% for both *B. calyciflorus* and *B. plicatilis* tests. The 24-h LC_{50} value for *B. calyciflorus* and *B. plicatilis* were 11850 µg/L and 10670 µg/L, respectively. These values were not used because essential information, such as percent active ingredient, was lacking.

Holbrook (1982) - In 1982, 24-h static toxicity tests were performed by the U.S. Department of Agriculture on technical grade chlorpyrifos (percent active ingredient not specified) with fourth instar larvae of ceratopogonid, *Culicoides variipennis*. Commonly recognized testing guidelines were not mentioned. Four concentrations of chlorpyrifos were tested and a dilution water control was used. Two replicates per concentration were tested. Measurement of chlorpyrifos concentrations was not mentioned. Water quality parameters during the test averaged: temperature of 22 $\pm$1^{o}C; pH of 7.0; and dissolved oxygen was not measured. Control survival was not mentioned. The 24-h LC_{50} value for *C. variipennis* was 4.3 µg/L. This value was not used because test duration should have been 96-h (ASTM 1988a) and information for several test parameters were missing.

Holbrook (1983) - In 1983, 24-h static toxicity tests were performed by the U.S. Department of Agriculture on technical grade chlorpyrifos (percent active ingredient not specified) with larvae of ceratopogonid, *Culicoides variipennis*. Commonly recognized testing guidelines were not mentioned. Five concentrations of chlorpyrifos were tested and a dilution water control was used. Three replicates per concentration were tested. All concentrations were nominal. Water quality parameters during the test averaged: temperature of 22 $\pm 1^oC$; neutral pH; dissolved oxygen was not measured but larvae remained close to the surface; and soft water was used. Control survival was greater than 90%. The 24-h LC_{50} value for *C. variipennis* was 2.96 µg/L. This value was not used because test duration should have been 96-h (ASTM 1988a) and information for several test parameters were missing.

<u>Kenaga et al. (1965)</u> - In 1965, 24-h toxicity tests were performed by the Dow Chemical Company on Dursban^R (percent active ingredient not specified) with adult and nymph cladoceran *Daphnia sp.*, immature goldfish *Carassius auratus*, and mature snail *Helisoma trivolvis*. Chemical Specialties Manufacturers Association (1963) testing guidelines were followed. Three concentrations of Dursban^R were tested. There was no mention of the use of controls. The number of replicates tested was not mentioned. Measurement of chlorpyrifos concentrations was not mentioned. Water quality parameters during the test averaged: temperature of 65^oF; pH was not mentioned; dissolved oxygen level was not mentioned; and hardness was not mentioned. Control survival was not mentioned. The 24-h LC_{50} value for cladoceran, goldfish, and snail were 16 µg/L, 180 µg/L, and >2000 µg/L, respectively. These values were not used because acute toxicity tests using fish must be 96-h in duration (ASTM 1988a), and essential information, such as the use of controls and control survival, was lacking.

<u>Sanders (1972)</u> - In 1972, 96-h static toxicity tests were performed by the U.S. Bureau of Sport Fisheries and Wildlife on technical grade Dursban^R (97%) with amphipod *Gammarus fasciatus*. Commonly recognized testing guidelines were not mentioned. Five concentrations of Dursban^R were tested. The use of a control was not mentioned. One replicate per concentration was tested. Measurement of chlorpyrifos concentrations was not mentioned. Water quality parameters during the test averaged: temperature of 21 $\pm 0.5^oC$; pH of 7.1; dissolved oxygen level of 8 mg/L; and alkalinity of 35 mg/L. Control survival was not mentioned. The 96-h LC_{50} value for *G. fasciatus* was 0.32 µg/L. This value was not used because essential information, such as the use of controls and control survival, was lacking.

<u>Schimmel et al. (1983)</u> - In 1983, 96-h flow-through toxicity tests were performed by the U.S. EPA on technical grade chlorpyrifos (92%) with Atlantic silverside *Menidia menidia*, mysid *Mysidopsis bahia*, sheepshead minnow *Cyprinodon variegatus*, longnose killifish *Fundulus similis*, and striped mullet *Mugil cephalus*. ASTM (1980) testing guidelines were followed. Chlorpyrifos concentrations tested were not mentioned. Solvent and dilution water controls were used. Three replicates per concentration were tested in mysid tests. The number of replicates tested in fish tests was not mentioned. Chlorpyrifos concentrations were measured but values were not given. Water quality parameters during the test were not mentioned. Control survival was not mentioned. The 96-h LC_{50} value for the Atlantic silverside, mysid, sheepshead minnow,

longnose killifish, and striped mullet were 1.7 µg/L, 0.035 µg/L, 136 µg/L, 4.1 µg/L, and 5.4 µg/L, respectively. These values were not used because essential information, such as water quality parameters and control survival, was lacking.

Strickman (1985) - In 1985, 7-d static toxicity tests were performed by the US Air Force Occupational and Environmental Health Laboratory on technical grade chlorpyrifos (93 to 100%) with second instar mosquito *Wyeomyia smithii*. Commonly recognized testing guidelines were not mentioned. Three concentrations of chlorpyrifos were tested and a solvent control was used. Eight replicates per concentration were tested. Measurement of chlorpyrifos concentrations was not mentioned. Water quality parameters during the test averaged: temperature of 27^{o}C; pH was not mentioned; dissolved oxygen level was not mentioned; hardness was not mentioned. Control survival was not mentioned. The data from this study were not used because essential information, such as LC_{50} value, control survival, and dissolved oxygen level, was lacking.

Thirugnanum and Forgash (1977) - In 1977, 96-h flow-through toxicity tests were performed by the Department of Entomology and Economic Zoology, New Brunswick State University on technical grade chlorpyrifos (99.5%) with mummichog *Fundulus heteroclitus*. Commonly recognized testing guidelines were not mentioned. Five concentrations of chlorpyrifos were tested and a solvent control was used. Two replicates per concentration were tested. Measurement of chlorpyrifos concentrations was not mentioned. Water quality parameters during the test averaged: temperature of 25^{o}C; pH of 7.5 to 8.0; dissolved oxygen level was not mentioned but the testing solution was aerated; and salinity of $20^{o}/_{oo}$ to $25^{o}/_{oo}$. Control survival was not mentioned. The 96-h LC_{50} value for *F. heteroclitis* was 4.65 µg/L. The 96-h LC_{50} value for *F. heteroclitis* was not used because essential information, such as control survival, was lacking.

U.S. Army Environmental Hygiene Agency (1970) - In 1970, toxicity tests were performed by the U.S. Army Environmental Hygiene Agency on Dursban^{R} (percent active ingredient not specified) with first instar larvae of mosquito *Culicoides pipiens quinquefasciatus*. Commonly recognized testing guidelines were not mentioned. Eight concentrations of Dursban^{R} were tested and a solvent control was used. Four replicates per concentration were tested. Measurement of chlorpyrifos concentrations was not mentioned. Water quality parameters during the test were not mentioned. Control survival was not mentioned. The test found that sublethal concentrations of Dursban^{R} had no effect on larval development of *C. pipiens quinquefasciatus*. This information was not used because essential information, such as acute toxicity data and control survival, was lacking.

Walton et al. (1990) - In 1990, 24-h static toxicity tests were performed by the Department of Entomology, University of California at Riverside on chlorpyrifos (the percent active ingredient not specified) with 4-5 day old tadpole shrimp *Triops longicaudatus*. Commonly recognized testing guidelines were not mentioned. Measurement of chlorpyrifos concentrations was not mentioned. A solvent control was used. Ten replicates per concentration were tested. Water quality parameters during the test averaged: temperature of 28 $\pm 2^{o}$C; pH was not mentioned; dissolved oxygen level was not mentioned. Control survival was not mentioned. The 24-h LC_{50} value for *T. longicaudatus* was 4.0 µg/L. This value was not used because essential information, such as concentrations tested and control survival, was lacking.

Accepted chronic toxicity tests - The following tests for chlorpyrifos evaluated in Menconi and Paul (1994) used accepted test methods.

Cripe et al. (1986) - In 1986, 28-d flow-through chronic toxicity tests were performed by the U.S. EPA on technical grade chlorpyrifos (92%) with sheepshead minnow *Cyprinodon variegatus*. Commonly recognized chronic test guidelines were not mentioned. Five concentrations were tested and a solvent control was used. Two replicates per concentration were tested. Chlorpyrifos concentrations were measured weekly and averaged 66 to 76% of nominal concentrations for the first series of tests and 64 to 74% of nominal concentrations for the second series of tests. Water quality parameters during the test averaged: temperature of 30 $\pm 1^oC$; pH range of 7.9 to 8.1; dissolved oxygen level of 5.0 mg/L; and salinity of $28^o/_{oo}$ for the first series of tests and $25^o/_{oo}$ for the second series of tests. Control survival was 97 to 100% for the first series of tests and 96 to 100% for the second series of tests. The NOEC value and LOEC value, based on growth, for *C. variegatus* were 1.7 µg/L and 3.0 µg/L, respectively. The MATC value was 2.26.

Goodman et al. (1985a) - In 1985, 28-d flow-through toxicity tests were performed by the U.S. EPA on technical grade chlorpyrifos (97.7%) with 32 to 36-h old embryos of inland silverside *Menidia beryllina* and tidewater silverside *Menidia peninsulae*. Commonly recognized testing guidelines were not mentioned. Five concentrations of chlorpyrifos were tested and solvent and dilution water controls were used. One replicate per concentration was tested. Chlorpyrifos concentrations were measured weekly and averaged 63 to 90% of nominal concentrations in *M. beryllina* tests and 65 to 78% in *M. peninsulae* tests. Water quality parameters during the test averaged: temperature of 25 $\pm 2^oC$; pH was not measured but was estimated to range from 7.3 to 8.1; dissolved oxygen level of 7.6 mg/L to 7.8 mg/L for *M. beryllina* and 6.0 mg/L to 6.9 mg/L for *M. peninsulae*; and salinity of $4.0^o/_{oo}$ to $6.0^o/_{oo}$ for *M. beryllina* and $18^o/_{oo}$ to $25^o/_{oo}$ for *M. peninsulae*. Sea water and solvent control survival for *M. beryllina* were 80% and 83%, respectively. Sea water and solvent control survival for *M. peninsulae* were 88% and 63%, respectively. NOEC values, based on growth, for *M. beryllina* and *M. peninsulae* were 0.75 µg/L and 0.38 µg/L, respectively. The LOEC values based on growth, for *M. beryllina* and *M. peninsulae* were 1.8 µg/L and 0.78 µg/L, respectively. The MATC values for *M. beryllina* and *M. peninsulae* were 1.16 and 0.54, respectively. This test also included Atlantic silverside *Menidia menidia,* described in ***Unaccepted chronic toxicity tests***.

Hansen et al. (1986) - In 1986, a 49-d flow-through toxicity test was performed by the U.S. EPA on technical chlorpyrifos (92%) with gulf toadfish *Opsanus beta*. ASTM (1985) testing guidelines were followed. Six concentrations of chlorpyrifos were tested and a solvent control was used. Two to three replicates per concentration were tested. Chlorpyrifos concentrations were measured weekly and averaged 50 to 60% of nominal concentrations. Water quality parameters during the test averaged: temperature of 26 $\pm 2^oC$; pH was not mentioned; dissolved oxygen level of 4.1 mg/L to 6.4 mg/L; and salinity of $25^o/_{oo}$ to $34.5^o/_{oo}$. Control survival was

97% The NOEC value and LOEC value, based on growth, were 1.4 µg/L and 3.7 µg/L, respectively. The MATC value for *O. beta* was 2.28.
Jarvinen and Tanner (1982) - In 1982, 32-d flow-through toxicity tests were performed by the U.S. EPA on technical grade chlorpyrifos (98.7%) with <48-h embryos of fathead minnow *Pimephales promelas*. APHA (1975) testing guidelines were followed. Five concentrations of chlorpyrifos were tested and a dilution water control was used. Two replicates per concentration were tested. Chlorpyrifos concentrations were measured weekly and averaged 102 to 144% of nominal concentrations. Water quality parameters during the test averaged: temperature of 25.0 $\pm$0.6^{o}C; pH of 7.4 to 7.8; dissolved oxygen level of 6.5 mg/L to 8.4 mg/L; hardness of 45.8 mg/L; and alkalinity of 43.1 mg/L. Control survival was 100%. The NOEC value and LOEC value, based on survival, for *P. promelas* were 1.6 µg/L and 3.2 µg/L, respectively. The MATC value for *P. promelas* was 2.26.

McKenney et al. (1981) - In 1981, 28-d flow-through toxicity tests were performed by the U.S. EPA on technical grade chlorpyrifos (97.7%) with juvenile mysid *Mysidopsis bahia*. Commonly recognized testing guidelines were not mentioned. Four concentrations of chlorpyrifos were tested and a solvent control was used. Eight replicates per concentration were tested. Chlorpyrifos concentrations were measured weekly and averaged 71 to 120% of nominal concentrations. Water quality parameters during the test averaged: temperature of 25 $\pm$2^{o}C; pH was not measured; dissolved oxygen level of 6.6 $\pm$0.2 mg/L; and salinity of 19$^{o}/_{oo}$ to 28$^{o}/_{oo}$. Control survival was 74%. The NOEC value, LOEC value, and MATC value, based on growth, for *M. bahia* were 0.002 µg/L, 0.004, and 0.003 µg/L, respectively.

Norberg and Mount (1985) - In 1985, 7-d static toxicity tests were performed by U.S. EPA on technical Dursban^{R} (percent active ingredient not specified) with larval fathead minnow *Pimephales promelas*. EPA (1982) and ASTM (1983) testing guidelines were followed. Five concentrations were tested and a dilution water control was used. Three replicates per concentration were tested. Chlorpyrifos concentrations were measured and averaged 60 to 74% of nominal concentrations. Water quality parameters during the test averaged: temperature of 25 $\pm$2^{o}C; pH of 7.8 to 8.0; initial dissolved oxygen level of 8.0 mg/L to 6.0 mg/L at 24-h; and hardness of 45 mg/L to 48 mg/L as $CaCO_3$. Control survival was $\geq$80%. The 7-d NOEC value and LOEC value for *P. promelas* based on growth were 3.7 µg/L and 7.4 µg/L, respectively. The MATC value was 5.23.

Unaccepted chronic toxicity tests - The following tests for chlorpyrifos evaluated in Menconi and Paul (1994) did not use accepted test methods.

Goodman et al. (1985b) - In 1985, 26-d and 35-d flow-through toxicity tests were performed by the U.S. EPA on technical grade chlorpyrifos (92%) with 2.5-d old fry California grunion *Leuresthes tenuis*. Commonly recognized testing guidelines were not mentioned. Five concentrations of chlorpyrifos were tested and solvent and dilution water controls were used. Three replicates per concentration were tested. Chlorpyrifos concentrations were nominal. Water quality parameters during the test averaged: temperature of 23^{o}C to 26^{o}C; pH of 7.6 to 7.9; dissolved oxygen level of 5.7 mg/L to 5.8 mg/L; and salinity of 28.6$^o/_{oo}$ in the 35-d tests and 29.3$^o/_{oo}$ in the 29-d tests. Control survival was 85% in the 26-d toxicity test and 78 to 82% in the 35-d toxicity test. The 26-d NOEC value and LOEC values for *L. tenuis* were 0.50 µg/L and 1.0 µg/L, respectively. The 35-d NOEC value and LOEC value for *L. tenuis* were 0.25 µg/L and 0.50 µg/L, respectively. These values were not used because chlorpyrifos concentrations were nominal and 2.5-d old fry were used instead of <48-h old embryos.

Goodman et al. (1985a) - In 1985, 28-d flow-through toxicity tests were performed by the U.S. EPA on technical grade chlorpyrifos (92%) with 32 to 36-h old embryos of Atlantic silverside *Menidia menidia*. Commonly recognized testing guidelines were not mentioned. Five concentrations of chlorpyrifos were tested and a solvent control was used. One replicate per concentration was tested. Chlorpyrifos concentrations were measured weekly and averaged 48 to 132% of nominal concentrations. Water quality parameters during the test averaged: temperature of 25 $\pm$2^{o}C; pH was not mentioned; dissolved oxygen level of 5.2 mg/L to 5.5 mg/L; and a salinity of 18$^o/_{oo}$ to 27$^o/_{oo}$. Control survival was 41%. The NOEC value and LOEC value for *M. menidia* were 0.28 µg/L and 0.48 µg/L, respectively. These data were not used because control survival in chronic toxicity tests must be greater than 60% (ASTM 1988c). The *M. menidia* tests had a control survival of 41%. This test also included tests using inland silverside *Menidia beryllina* and tidewater silverside *Menidia peninsulae*. These tests were acceptable and are described in ***Acceptable chronic toxicity test abstracts***.

Kersting and Van Wijngaarden (1992) - In 1992, 21-d static toxicity tests were performed by the Research Institute for Nature Management, Netherlands on technical grade chlorpyrifos (99%) with <24-h old cladoceran *Daphnia magna*. Commonly recognized testing guidelines were not mentioned. Six concentrations of chlorpyrifos were tested and solvent and dilution water controls were used. Two replicates per concentration were tested. Chlorpyrifos concentrations were measured at the beginning of each test and averaged 60% of nominal concentrations. Water quality parameters during the test averaged: temperature of 19.5 $\pm$0.5^{o}C; pH of 6.8 to 7.0; and dissolved oxygen level of 7.7 mg/L to 8.8 mg/L. Control survival was 100%. The NOEC value, based on reproduction, for *D. magna* was 0.1 µg/L. These values were not used because a LOEC value was not determined and concentrations were not measured during the test.