060101
SHAUGHNESSEY NO.

REVIEW NO.

EEB BRANCH REVIEW

DATE: IN 2/11/82 OUT 5/28/82

FILE OR REG. NO. 618-75

PETITION OR EXP.

DATE OF SUBMISSION January 8, 1982

DATE RECEIVED BY HED February 9, 1982

RD REQUESTED COMPLETION DATE May 29, 1982

EEB ESTIMATED COMPLETION DATE

RD ACTION CODE/TYPE OF REVIEW 400/Data Submission

TYPE PRODUCT(S): I, D, H, F, N, R, S Fungicide

DATA ACCESSION NO(S).

PRODUCT MANAGER NO. H. Jacoby (21)

PRODUCT NAME(S) Mertect 340-F

COMPANY NAME Merck Sharp & Dohme Research Laboratories

SUBMISSION PURPOSE Submission of Data to Support Proposed Use on Rice

SHAUGHNESSEY NO.	CHEMICAL & FORMULATION	% A.I.
060101	2-(4-thiazolyl)benzimidazole	42.28

2019496

100.0 Purpose of Submission

Validation of toxicity studies required to support the conditional registration of Mertect 340 F fungicide on rice.

103.0 Toxicological Properties

Chemical: Mertect 340

Formulation: Technical (98.5% active ingredient)

Test Type: Embryo-Larvae (EPA Acc# 246711)

Test Organism: Fathead minnow (Pimephales promelas)

Results: Maximum acceptable toxicant concentration (MATC) = > 0.05 < 0.97 mg/l

typo: should be > 0.5 < 0.97 mg/l see attached sheet

Validation Status: Core

Comments: Results from this study can be used to support product registration.

Chemical: Mertect 340F

Formulation: Technical (98% active ingredient)

Test type: Chronic freshwater invertebrate

Test organism: Daphnia magna

Results: Maximum acceptable toxicant concentration (MATC) = > 0.042 < 0.087 mg/l

Validation Status: Core

Comments: Results from this study can be used to support product registration.

104.0. Discussion

Mertect 340F (EPA Reg. 618-75) was conditionally registered for use on rice in June of 1980. Fish and wildlife studies needed to support conditional registration are shown below (Bowen, 6/5/80):

1. An aquatic invertebrate life-cycle test.
2. Fish embryo-larvae test using rainbow trout and the fathead minnow.

2

The Registrant (Merteck and Co.) was also asked to modify the product label to include the statement "This pesticide is toxic to fish."

The rainbow trout embryo-larvae study (EPA. Acc# 247102) requested by EEB has been submitted by the registrant and is currently being validated by our Branch. EEB is satisfied with the registrant's new product label (Appendix I) since it now contains precautionary statements that will help minimize Mertect's impact on non-tagret fish and aquatic invertebrates. The Ecological Effects Branch will integrate all new data into an incremental risk assessment for non-target fish and invertebrates following the validation of the rainbow trout toxicity study.

107.0 Conclusions

107.2 Data Adequacy

The following bioassays have been classified as acceptable and can be used in support of the conditional registration of Mertect 340F on rice:

1. The toxicity of Mertect fungicide to fathead minnow (Pimephales promelas) embryo and larvae (EPA Acc # 246711).

2. The chronic toxicity of Mertect fungicide to the water flea, Daphnia magna (EPA Acc # 246711).

107.5 Recommendations

EEB will complete an incremental risk assessment [3(c)(7) Finding] for this conditionally registered fungicide following the validation of the rainbow trout toxicity study (EPA ACC # 247102).

Charles A. Bowen II
Fisheriers Biologist, Section I
Ecological Effects Branch
Hazard Evaluation Division (TS-769) Date: 5/28/82

Raymond W. Matheny, Head
Section I
Ecological Effects Branch
Hazard Evaluation Division (TS-769) Date: 5/28/82

Clayton Bushong, Chief
Ecological Effects Branch
Hazard Evaluation Division (YS-769) Date: 5/28/82

Chemical: Thiabendazole

Formulation: Technical (98.5% activie ingredient)

Citation: Sorprenant, D.C. 1981. The chronic toxicity of Mertect fungicide to the water flea (Daphnia magna). EG&G, Bionomics Aquatic Toxicology Laboratory. Wareham, MA.

Reviewed By: Charles A. Bowen II, Fisheries Biologist
Ecological Effects Branch
Hazard Evaluation Division (TS-769)

Test Type: 48-hour freshwater invertebrate bioassay.

A. Test organism: Daphnia magna

Reported Results: Survival of daphnids was significantly affected at Mertect fungicide exposure concentrations as low as 0.087 mg/l. Offspring production was significantly reduced among daphnids exposed to 0.087 mg/l. Based on these data, the minimum threshold concentration (MTC) of this compound for D. magna was >0.042 <0.087 mg/l.

Reviewer's Conclusion: This bioassay is scientifically sound and demonstrates both effect and no effect levels for the on fathead minnow. Data from this bioassay can be used to support product registration.

Materials and Materials:

The methodology for conducting this study was based on "Methods for acute toxicity tests with fish, macroinvertebrates, and amphibians" (U.S. EPA, 1975) and "Protocol for conducting chronic toxicity tests with the water flea (Daphnia magna)" developed at EG&G, Bionomics (1980). The selection of concentrations of Mertect fungicide for the chronic toxicity test were based on the results of the static acute toxicity test also conducted at EG&G, Bionomics for the Merck & Company, Inc.

Diluent water used in the acute and chronic toxicity tests consisted of deionized well water, reconstituted to a total hardness of 170 $\pm$ 20 mg/l as $CaCO_3$, a pH of 7.9-8.3, a specific conductance of 500 $\pm$ 100 umhos/cm and a total alkalinity of 120 $\pm$ 10 mg/l as $CaCO_3$. The water was reconstituted in a 1800-l fiberglass tank and pumped to the test system with an FMI Model RPD laaboratory metering pump.

Acute Procedure

Each test concentration of Mertect fungicide used during the acute toxicity test was prepared by adding the appropriate volume of test material, dissolved in DMF, into 0.5 l of water and vigorously mixing the solution on a magnetic stirrer. Fifteen daphnids ($\leq$24 hours old) were subsequently added to each solution. A control, consisting of the same dilution wate rand conditions, but containing no Mertect fungicide, was maintained during the test. In addition, a solvent control containing the greatest concentration of DMF present in any test vessel (0.5 mg/l), was also maintained. Mortality fo the test organisms was assessed at 24 and 48 hours of exposure. The test was maintained at 21 $\pm$ 1°C by ambient room temperature. Mortality data were used to calculate median lethal concentration (LC_{50}) values at each 24 hour interval. The LC_{50} is defined as the concentration (nominal or measured) of the test compound in dilution water which casued mortlaity of 50% of the test animal population. The LC_{50} calculations were based on nominal concentrations of Mertect fungicide.

Chronic Procedure

Aquaria were glass battery jars having a volume capacity of 1.75 liters. Test solutions drained from aquaria through a 3 x 8 cm notch cut on the upper edge of the jars. Notches were covered with a Nitex® 40 mesh screen to prevent loss of the daphnids. Five concentrations of Mertect fungicide were assessed in addition to two separate sets of controls. One control group (solvent control0 received the greatest amount of DMF present in any treatment (42 ul/l). All treatments and controls were conducted in guadruplicate. Test solutions were delivered to the aquaria at a rate of four aquarium volumes per day.

The dissolved oxygen contration and temperature of the test solutions were monitored on every weekday within replicate aquarium of each treatment level and controls. Total hardness, alkalinity, specific conductance and pH of the test solutions were monitored weekly in one aquarium from each treatment and controls. Dissolved oxygen concentrations were determined with a YSI Model 54BP dissolved oxygen meter. Temperatures were measured with a Weston dial thermometer. Specific conductances were monitored with a YSI Model 33 conductivity-salinity-meter. The pH of the test solutons were determined with an Instrumentation Laboratories Model 175 pH meter. Total hardness and alkalinity of test solutions were determined according to APHA et al. (1975).

One hundred milliliter water samples were removed from two replicate test vessels of the high, middle and low test concentrations and control on two days prior to initiating the chronic test. Results of these analyses were used to judge whether sufficient quantities of Mertect fungicide were being delivered to the aquaria to initiate the test. During the exposure, one hundred milliliter water samples were removed weekly from two replicate test vessels of each treatment level and controls for the determination of Mertect fungicide concentrations. Two quality assurance samples were prepared at each sampling interval during the exposure and remained with the set of samples through extraction and analysis. These samples were prepared in diluent water at a Mertect fungicide concentration unknown to the analyst. Each Adult survival was determined weekly and determinations of offspring production were made on every weekday from day 7 through 21. The offspring were removed, conted with a Fisher Count-All® Model 600 particle counter (LeBlanc, 1979) and discarded.

Chronic test organisms were fed a combination of PR-11® fish food suspension (5 mg/ml) and unicellular green algae (Ankistrodesmus sp.) (7.5×10^7 cells/ml). The food was introduced at a rate of 2 ml of PR-11® suspension and 1 ml of algal suspension three times daily on weekdays and once daily on weekends. Test aquaria were rinsed with diluent water, followed by the replacement of the original test solution, whenever survival and reproduction were assessed.

Statistics

The acute LC_{50} values were calculated using a computer program (Stephan, 1978, personal communication) which estimated the LC_{50} values using one of three statistical methods in the following order of preference: moving average angle analysis, probit analysis, or binomial probability. The methods selected were determined by the characteristics of the data base (i.e, presence or absence of 100% response, number of partial responses, etc.). The computer program scanned the data base, identified the most preferred statistical method and performed the analysis.

Weekly survival data, transformed to arc sin percentage and the determinations of cumulative production of offspring per female derived during the chronic toxicity test, were subjected to analysis of variance according to Steel and Torrie (1960). If significant (P=0.05) differences between treatments were observed, the Dunnett's procedure (Steel and Torrie, 1960) was used to determine which treatments, if any, varied from the controls. Results of the statistical analyses were used to estimate the minimum threshold concentration (MTCO. The MTC is defined as the minimum amount of Mertect fungicide, in water, which elicited an adverse response by D. magna during the chronic toxicity test.

Results

Nominal concentrations of Mertect fungicide tested and corresponding percentage moralities of D. magna observed during the acute exposure are presented in Table 3. The 24-hour LC_{50} was empirically estimated to be >1.0 mg/l, the highest concentration tested. The 48-hour LC_{50} and corresponding 95% confidence interval, calculated by binomial probability, was 0.28(0.22-0.36) mg/l.

Chronic Toxicity Test

Results of preliminary analyses of three test concentrations of Mertect fungicide and the control indicated that the measured Mertect fungicide concentrations were close to nominal in the highest and lowest concentrations measured (84 and 81%, respectively) (Table 4). The pre-test #1 samples of the middle concentration (0.062 mg/l nominal) contained an average of 0.24 mg/l Mertect fungicide. The reason for the high measured concentration is presumed to be due to sampling error since 0.24 mg/l is comparable to the highest test concentration. The pre-test #2 samples at this treatment level averaged 0.053 mg/l Mertect fungicide which is 85% of nominal. Upon completion of the pre-test sampling and analyses, the test concentrations were adjusted to supply a high Mertect fungicide concentration of 0.50 mg/l. This measure was taken to insure the determination of the MTC. Analysis of test solutions during the chronic wxposure indicated the test solutions averaged from 68 to 91% of nominal (Table 5). Results of the water quality analyses of test solutions indicated that the dissolved oxygen concentrtion, temperature, total harness, alkalinity, specific conductance, and pH varied minimally within and between treatment levels throughout the exposure (Table 6).

All of the daphnids exposed to 0.087 Mertect fungicide was significantly reduced as compared to survival of control daphnids after 7 days of exposure. Reduced survival at this treatment level was signficant as compared to both control and solvent control daphnids by day 14 of the exposure period. No increased effects of Mertect fungicide on daphnid survival were observed between days 14 and 21.

Daphnids exposed to 0.087 mg/l Mertect fungicide produced no offspring until after day 10 of exposure (Figure 1). Daphnids exposed to 0.024, 0.042 mg/l Mertect fungicide, the control and solvent control were producing offspring by day 7 of exposure. The cumulative production of offspring by daphnids exposed to 0.087 mg/l Mertect fungicide was significantly less than the production of offspring by solvent control daphnids beyond day 8 and was significantly less than the production of offspring by both control and solvent control daphnid beyond day 15. Several daphnids exposed to 0.087 mg/l Mertect fungicide were observed carrying dead (white colored0 eggs in their brood chambers. In addition, several molted, exoskeletons contained dead eggs in this treatment level. The production of offspring by daphnids exposed to 0.024 and 0.042 mg/l Mertect fungicide was comparable to offspring production by control and solvent control daphnids for the duration of the exposure.

Based on the reduced survival and reproduction of daphnids exposed to Mertect fungicide concentrations as low as 0.087 mg/l, the MTC of this compound for D. magna was estimated to be >0.042 <0.087 mg/l.

References

APHA, AWWA, WPCF. 1975. Standard methods for the examination of water and wastewater. 14th Edition, New York, Hardness EDTA Titrimetric Method. 309B, 203-206.

LeBlanc, G.A. 1979. Utilization of bacterial colony counters to count early instar water flea (Daphnia magna). Bull. Environm. Contam. Toxicol. 23: 837-839.

Steel, R.G.D. and J.H. Torrie. 1960. Principles and procedures of statistics. McGraw-Hill, New York: 481 pp.

Stephan, Charles. 1978. U.S. EPA, Environmental Research Laboratory, Duluth, Minnesota. Personal communication.

U.S. EPA. 1975. Methods for acute toxicity tests with fish, macro-invertebrates, and amphibians. Ecological Research Series (EPA-660/3-75-009), 61 pp.

EEB Statisical Validation (21 days):

Highest No Effect level

	DEAD	ALIVE	
Test group (0.042 mg/l)	11	69	80
Solvent Control	10	70	80
	21	139	160

$$X^2 = \frac{[(11 \times 70) - (10 \times 69)]^2 \times 160}{21 \times 139 \times 80 \times 80}$$

$$X^2 = \frac{[770 - 690]^2 \times 160}{18,681,600}$$

$$X^2 = \frac{[80]^2 \times 160}{18,681,600}$$

$$X^2 = \frac{6,400 \times 160}{18,681,600}$$

$$X^2 = \frac{1,024,000}{18,681,600} = 0.0548$$

X^2 cutoff 1 degree freedom = 3.84

X^2 = 1df = $P < .05$; Hence no significant difference between 0.042 group and solvent control.

Significant Effect Level:

Test group (0.087 mg/l)	32	48	80
Solvent Control	10	70	80
$\leq$	42	118	160

$$X^2 = \frac{[(32 \times 70) - (48 \times 10)]^2 \times 160}{42 \times 118 \times 80 \times 80}$$

$$X^2 = \frac{(2,240 - 480)^2 \times 160}{31,718,400}$$

$$X^2 = \frac{3,097,600 \times 160}{31,718,400} = \frac{495,616,000}{31,718,400}$$

$$X^2 = 15.62$$

X^2 cutoff for 1 degree freedom = 3.84

P>.005; Hence chi-squared test indicates that significant difference exists between the 0.087 mg/l group and solvent control.

EEB Statistical Validation:

MATC values calculated by the above method do not differ significantly from the values reported by the author.

Validity of Author's Conclusions:

The conclusions drawn by the author are supported by dose related mortality data cited in Table 7.

Validation Status:

Core

Validation Rationale:

The methods employed by the registrant's testing facility adhere to EPA's guidelines for chronic freshwater invertebrate studies. The author's conclusion are supported by the dose related mortality cited in Table 7.

Category Repairability:

N/A

Table 1. Analytical results of Mertect fungicide recovery samples.

Nominal concentration (µg/mℓ)		Mass added (µg)	Mass recovered (µg)	% Recovery
1.2	A	120	120	100
	B	120	110	92
	C	120	120	100
0.12	A	12	9.6	80
	B	12	9.6	82
	C	12	12	100
0.012	A	1.2	1.0	83
	B	1.2	1.4	120
	C	1.2	1.5	120
Blank	A	N/A	not detected	N/A
	B	N/A	not detected	N/A
	C	N/A	not detected	N/A

Average Recovery 97 ± 15

Table 2. Analytical results for quality control blind samples prepared at each sampling period during the chronic exposure of D. magna to Mertect fungicide.

Test day	Expected concentration (µg/mℓ)	Measured concentration (µg/mℓ)	% Recovery
0	1.2	1.0	83
	2.4	2.3	96
7	0.60	0.44	73
	2.4	1.9	79
14	0.60	0.52	87
	1.2	0.95	79
21	1.2	0.97	81
	1.2	0.87	72

Table 3. Concentrations tested and corresponding observed percentage mortalities for the water flea (Daphnia magna) acutely exposed to Mertect fungicide.

Nominal concentration (mg/ℓ)	Percentage mortality	
	24 hour	48 hour
1.0	7[a]	100
0.60	0	100
0.36	0	80[a]
0.22	0	0[a]
0.13	0	0
solvent control	0	0
control	0	0

[a] Several daphnids were lethargic.

Table 4. Mean (standard deviation) Mertect fungicide concentrations measured before initiating the chronic exposure of D. magna.

Nominal concentration (mg/ℓ)	Measured concentration (mg/ℓ)
0.25	0.21(0.01)
0.062	0.15(0.11)
0.016	0.013(0.003)
solvent control	<0.0068
control	<0.0068

Table 5. Mean (standard deviation) concentrations of Mertect fungicide measured during the chronic exposure of the water flea (Daphnia magna).

Nominal concentration (mg/ℓ)	Mean measured[a] concentration (mg/ℓ)
control	<0.0058
solvent control	<0.0062
0.031	0.024(0.011)
0.062	0.042(0.010)
0.12	0.087(0.016)
0.25	0.24(0.05)
0.50	0.39(0.07)

[a] n=8.

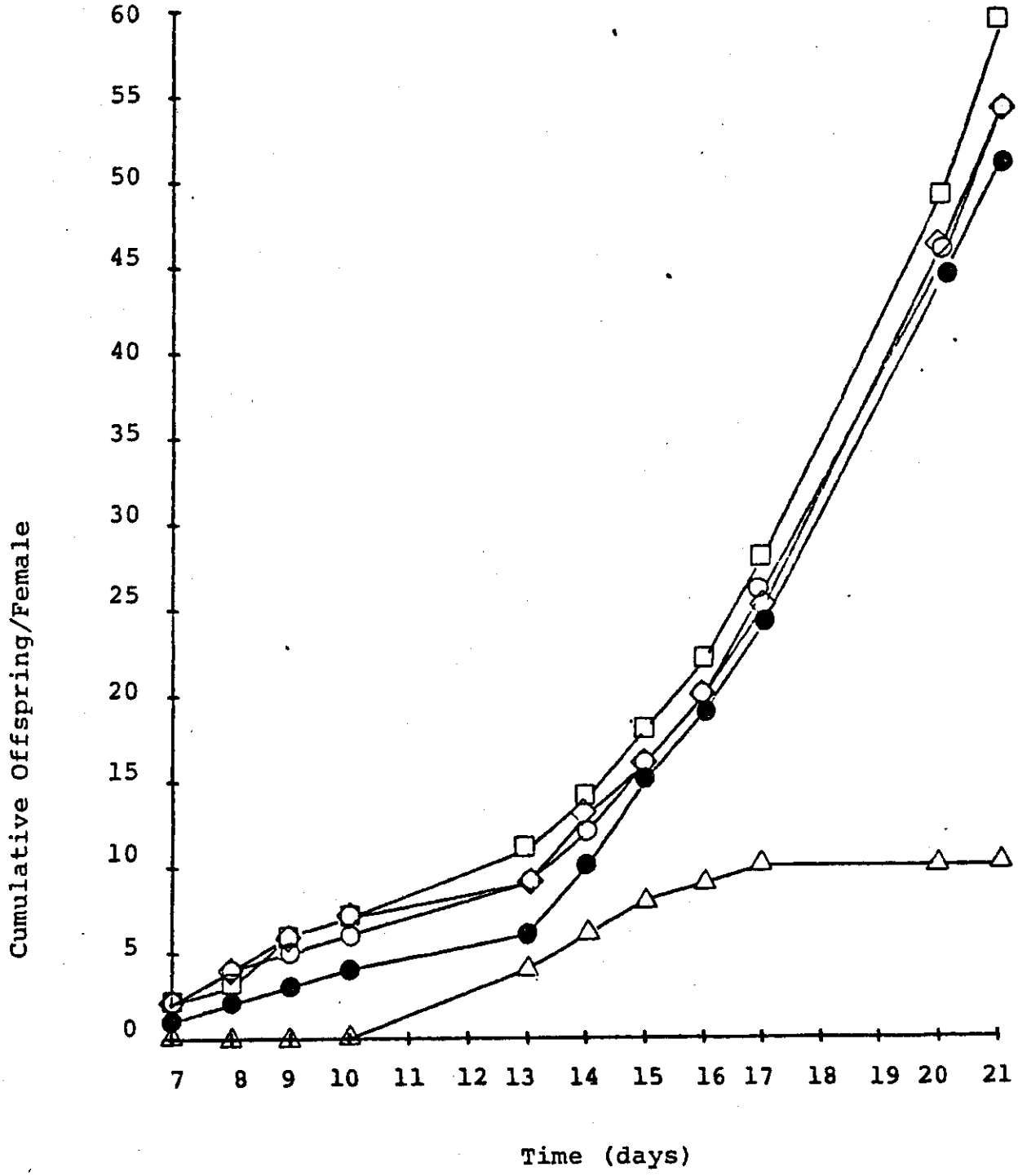
Table 6. Water quality analysis of test solutions during the chronic exposure of the water flea (Daphnia magna) to Mertect fungicide.

Mean measured concentration (mg/ℓ)	Mean (standard deviation)					
	Dissolved oxygen (mg/ℓ)	Temperature (°C)	Total hardness (mg/ℓ $CaCO_3$)	Total alkalinity (mg/ℓ $CaCO_3$)	Specific conductance (µmhos/cm)	pH range
control	7.8(0.3)	22(0)	180(0)	130(0)	500(0)	8.0-8.1
solvent control	7.8(0.3)	22(0)	180(0)	130(0)	500(0)	8.0-8.1
0.024	7.8(0.3)	22(0)	180(0)	130(0)	500(0)	8.0-8.1
0.042	7.8(0.4)	22(0)	180(0)	130(0)	500(0)	8.0-8.1
0.087	7.8(0.4)	22(0)	180(0)	130(0)	500(0)	8.0-8.1
0.24	8.1(0.2)	22(0)	180(0)	130(0)	500(0)	8.0-8.1
0.39	8.0(0.2)	22(0)	180(0)	130(0)	500(0)	8.0-8.1

Table 7. Weekly mean (standard deviation) percentage survival of the water flea (Daphnia magna) during chronic exposure to concentrations of Mertect fungicide.

Mean meaured concentration (mg/ℓ)	Day/ Percentage survival 7	14	21
control	99(2)	94(5)	89(8)
solvent control	98(3)	91(5)	88(3)
0.024	99(2)	91(11)	91(11)
0.042	99(2)	88(9)	86(11)
0.087	90(9)	69(15)	60(11)
0.24	0(0)	0(0)	0(0)
0.39	0(0)	0(0)	0(0)

Figure 1. Cumulative offspring produced per female D. magna during continuous exposure to concentrations of Mertect fungicide. ● = control, o = solvent control, □ = 0.024 mg/ℓ, ◇ = 0.042 mg/ℓ, △ = 0.087 mg/ℓ.



CHEMICAL: Thiabendazole

FORMULATION: Technical (98.5% Active ingredient)

CITATION: Wilson, B.F., 1981. The toxicity of Merteck fungicide to fathead minnow Pimephales promelas embryo and larvae. EG and G Bionomics Aquatic Toxicology Laboratory. Wareham, Massachusetts.

REVIEWED BY: Charles A. Bowen II
Fisheries Biologist
Ecological Effects Branch
Hazard Evaluation Division (TS-769)

DATE REVIEWED: May 17, 1982

TEST TYPE: 30-day embryo-larvae study

TEST ORGANISM: Fathead minnow (Pimephales promelas)

AUTHOR'S RESULTS:

Embryo hatchability was the most sensitive indicator of the effect of Mertect fungicide on the fathead minnow. Hatchability of embryos exposed to a mean measured concentration of 0.97 mg/l Mertect fungicide was significantly (P=0.05) reduced as compared to hatchability of control and solvent control embryos. No adverse effects on embryo hatchability or survival and growth of larvae from exposure to mean measured concentrations <0.05 mg/l Mertect fungicide were observed. The maximum acceptable toxicant concentration (MATC) of Mertect fungicide for fathead minnow embryos and larvae was estmated to be >0.050<0.97 mg/l.

REVIEWER'S CONCLUSION:

This bioassay is scientifically sound and has determined both effect and no-effect concentrations for the fathead minnow. Data from this study can be used to support product registration.

METHODS & MATERIALS:

The embryo and larvae test was conduced according to the protocol entitled "Methods for conducting early life stage toxicity tests with fathead minnow (Pimephales promelas)," prepared by EG&G, Bionomics. This test was conducted at the Aquatic Toxicology Laboratory of EG&G, Bionomics, Wareham, Massachusetts. Data generated during this study are stored at the above location.

A modified, proportional diluter, similar to that described by Mount and Brungs (1967) with a 0.50 dilution factor was used to deliver the desired concentration of Mertect fungicide to the aquaria during the embryo and larvae oxposure. The water was heated to 25 + 1°C in a gas fired glass coil heater and passed through a Rain Soft® water hardener containing calcite prior to delivery to the test system. The calcite increased the hardness and pH of the water to approximately 32 mg/l as calcium carbonate ($CaCO_3$) and 7.5, respectively.

Embryo and Larvae Exposure

The exposure of fathead minnow to Mertect fungicide was initiated with embryos obtained within 48 hours after fertilization from the harhead minnow culture unit at EG&G, Bionomics, Wareham, Massachusetts. Sixty embryos were impartially distributed to each of 14 embryo cups, one of which was suspended in each of the test aquaria. Embryo incubation cups were glass jars (5 cm O.D., 8 cm high) with 40 mesh Nitex® screen bottons. A rocker arm apparatus, as described by Mount (1968), was used to gently oscillate the incubation cups in the test water. Dead embryos were counted and removed daily until hatching was complete. Percentage successful hatch calculations were based on the number of live larvae per incubation cup after hatching was completed compared to the number of embryos per cup (60) at the initiation of exposure.

The initiate the 30-day larvae exposure, 40 larvae were impartially selected from each incubation cup and transferrred to the respective aquaria upon completion of hatching. Larvae were fed live brine shrimp (Artemia salina) nauplii three times daily on week days and twice daily on weekends and holidays. Aquaria were brushed and siphoned at least twice each week or as necessary to remove excess food and fecal matter. Behavior and appearance of larvae were observed daily and larvae were counted twice weekly. At 30 days post-hatch, the larvae from each aquarium were anesthetized with MS-222 (tricaine methanesulfonate) and percentage survival, mean total length and average wet weight were determined. The larvae were measured individually to calculate mean and standard deviation total length while the mean body weight for larvae in each aquarium was calculated from the total wet weight of all individuals in the aquarium. The study was initiated on 18 March and ended on 22 April 1981.

Statistics

Percentage hatch of embryos and survival, length and weight of larvae after 30-days exposure, were subjected to analysis of variance (Steel and Torrie, 1960, completely randomized block design, P=0.05). Data for percentage hatch and percentage survival were transformed to arc sin percentage prior to analysis. If treatment effects were indicated, the means of these parameters were compared to those from the control and solvent control using Dunnett's procedure (Steel and Torrie, 1960). When a treatment mean was significantly different from the control means (P=0.05), that treatment was considered to be an effect level. Based on these data, the MATC of Mertect fungicide to fathead minnow embryos and larvae was determined.

RESULTS

Results of water quality analyses indicated that the temperature of the test solutions was maintained at 25 $\pm$ 1°C. The pH ranged from 7.2-8.0, the dissolved oxygen concentration ranged form 6.9-9.1 mg/l and total hardness as $CaCO_3$ ranged from 32-34 mg/l.

Analysis of the quality control blind samples prepared with each set of water samples indicated that considerable variablility existed in the percentage recovery of Mertect fungicide from the samples (Table 2). A Student's comparison of the percentage of Mertect fungicide recovered from the quality control blind samples and the highest Mertect fungicide test concentration measured at each sampling interval indicated that the trend in recovered Metect fungicide was not significantly different (P=0.05, t=0.46). This suggests that one source of the variability occurring between sets of samples was related to sampel preparation (i.e, extraction, storage, etc.) and can be corrected for at each sampling interval by dividing the measured Mertect fungicide concentrations by the mean percentage recovery of the quality control blind samples for that interval. Mean measured Mertect fungicide concentrations and corrected mean measured Mertect fungicide concentrations are presented in Table 3. Mean measured Mertect fungicide concentrations presented in these results are the corrected values.

Percentage successful hatch of fathead minnow embryos exposed to a mean measured concentration of 0.97 mg/l Mertect fungicide was significantly reduced as compared to percentage hatch of control and solvent control embryos (Table 4). Eighty three percent of the mortalities observed furing incubation were of unhatched embryos and 17% of the mortalities were of newly hatched embryos. Percentage hatch of embryos was unaffected by exposure to mean measured Mertect fungicide concentrations as high as 0.05 mg/l.

Survival data indicate that larvae exposed to 0.97 mg/l were affected in the A replicate while those in the B replicate were not. However, the number of larvae successfully hatched and transferred to the A replicate was about twice that of the B replicate, as reflected in the difference in percentage hatch for the two replicates. These larvae appeared lethargic at hatch and subsequently mortality during the first two days of larvae exposure reduced the number of surviving larvae in the A replicate to approximately that of the B replicate. Due to the variability between the replicate aquaria of the 0.97 mg/l treatment, the reduced survival was not statistically significant. Survival of larvae exposed to concentrations as high as 0.50 mg/l was comparable to survival of larvae in both control groups. Mean total length and average wet weight of larvae was unaffected by exposure to Mertect fungicide concentrations as high as 0.97 mg/l.

The larvae that had survived 30 days of exposure to a measured concentration of 0.97 mg/l Mertect fungicide had a greater average wet weight that control and solvent control larvae. This response has been observed in other partial and full life cycle studies when a reduction in the number of fish in an aquarium results in comparatively larger fish, presumably due to increased availability of food.

Based on the reduced percentage hatch of fathead minnow embryos exposed to 0.97 mg/l Mertect fungicide, the MATC of this compound for fathead minnow embryos and larvae was estimated to be >0.50 <0.97 mg/l.

REFERENCES

Mount, D.I. 1968. Chronic toxicity of copper to fathead minnow (Pimephales promelas). Water Res. 2: 215-223.

Mount, D.I. and W.A. Brungs. 1967. A simplified dosing apparatus for fish toxicology studies. Water Res. 1: 20-29.

Steel, R.G.D. and J.H. Torrie. 1960. Principles and procedures of statistics. McGraw-Hill, New York: 481 pp.

EEB Statistical Validation (Embryo):

	Dead Embryos	Alive Embryos	
.97 mg (group A)	29	31	60
Control B.	3	57	60
	32	88	120

$$X^2 = \frac{[(29X57)-(3X31)]^2 \times 120}{60 \times 60 \times 32 \times 88}$$

$$X^2 = (1,653-91)^2 \times 120$$

$$X^2 \quad \frac{(1562)^2 \times 120}{10,137,600} = \frac{292,781,280}{10,137,600} = 28.8$$

X^2 cutoff at 1 degree freedom = 7.88

Chi-square test indicated that the 0.97 concentration had a significant ($P > 0.001$) effect on embryo survival.

	Dead Embryos	Alive Embryos	
0.97 mg (B Group)	43	17	60
Control (B Group)	3	57	60
	46	74	120

$$X^2 = \frac{[(43 \times 57) - (17 \times 3)]^2 \times 120}{60 \times 60 \times 46 \times 76}$$

$$X^2 = \frac{[(2451 - 51)^2 \times 120}{691,200,000}$$

$$X^2 = \frac{(2400)^2 \times 120}{12,254,400} = 56.40$$

X^2 = 1df cutoff = 7.88 P = >0.001

Chi-square test indicates that the 0.97 mg/l concentration significantly (P > 0.001) effected developing fathead embryos.

EEB Statistical Conclusions:

MATC values calculated by the above method support the author's conclusions.

Validity of Authors Conclusions:

The conclusions drawn by the author are supported by the dose related mortality data cited in Table 2.

Validation Status: Core

Validation Rationale:

The methods employed by the registrant's testing facility adhere to EPA's guidelines for chronic embry-larvae studies. The author's conclusion are supported by the dose mortality data cited in Table 2.

Category Repairability: N/A

Table 2. Analytical results for quality control blind samples.

Test day	Expected concentration (µg/mℓ)	Measured concentration (µg/mℓ)	% Recovery
0	0.15	0.11	73
	0.15	0.12	80
5	0.75	0.45	60
	0.75	0.44	59
12	0.12	0.069	58
	0.12	0.083	69
19	0.60	0.53	88
	1.2	0.92	77
26	1.2	1.1	92
	2.4	2.2	92
34	0.60	0.47	78
	2.4	1.9	79

Table 3. Mean (standard deviation) concentrations of Mertect fungicide, corrected and uncorrected for low quality control blind samples measured with each sample set, measured during the exposure of fathead minnow (Pimephales promelas) embryos and larvae.

Nominal concentration (mg/ℓ)	Measured concentration (mg/ℓ)	Corrected measured concentration (mg/ℓ)
1.0	0.72(0.11)	0.97(0.15)
0.50	0.37(0.08)	0.50(0.10)
0.25	0.21(0.08)	0.29(0.11)
0.12	0.072(0.015)	0.10(0.02)
0.062	0.038(0.007)	0.052(0.011)
solvent control	<0.0045	<0.0058
control	<0.0078	<0.010

Table 4. Hatchability of embryos and survival, total length and wet weight of fathead minnow larvae exposed to Mertect fungicide.

Measured concentration (mg/ℓ)		60 Hatch (%)	40 Survival (%)	30 Day Old Larvae Total length (mm) $\bar{x}$ (± S.D.)[a]	Average weight (mg)
0.97	A	52[b]	58	23(2)	98
	B	28[b]	100	23(1)	94
0.50	A	95	100	21(2)	70
	B	97	100	21(2)	80
0.29	A	95	98	21(3)	70
	B	87	100	22(2)	82
0.10	A	93	95	22(2)	80
	B	95	100	22(2)	81
0.052	A	97	100	22(2)	78
	B	93	95	22(1)	81
solvent control	A	97	100	21(2)	73
	B	93	93	22(2)	83
control	A	95	100	22(2)	76
	B	95	98	22(1)	81

[a] Mean (and standard deviation).

[b] Mean of A and B replicates significantly (P=0.05) different from means control and solvent control replicates.

DIRECTIONS FOR USE

It is a violation of Federal (U.S.A.) law to use this product in a manner inconsistent with its labeling.

GENERAL—Shake or stir well before use.

Clean equipment before using MERTECT 340-F. When MERTECT 340-F is dispersed in water, the resulting suspension must be constantly agitated. Avoid foaming.

Compatibility information for MERTECT 340-F with other chemicals is limited. Consult your supplier or agricultural experiment station for specific recommendations.

SOYBEANS—**Reduce the Severity of Pod and Stem Blight, Anthracnose, Brown Spot, Frog Eye Leaf Spot and Purple Seed Stain (Ground and Aerial Applications):** Apply MERTECT 340-F at 6.0—10.0 fl. oz. per acre per application in sufficient water for coverage. Make two applications per season: the first application at late flowering to early pod set and the second application two weeks later. Make aerial applications in a minimum of 5 gallons of water per acre. Do not apply MERTECT 340-F within 21 days of harvest. Do not graze or feed treated soybean vines or hay to livestock. Do not tank mix with copper fungicides.

POME FRUIT (Apples and Pears)—**Control Blue Mold Rot, Bull's Eye Rot, and Gray Mold (Nest Rot, Sister Rot), Stem End and Neck Rot:** Dip, flood, or spray harvested fruit with a suspension of 16 fl. oz. MERTECT 340-F in 100 gal. water. Do not treat for over 3 minutes. Treat apples only before and after storage for maximum decay control. Treat pears once.

SUGAR BEETS—**Control Cercospora Leaf Spot:** Use 6—12 fl. oz. MERTECT 340-F per acre. GROUND Spraying—Use MERTECT 340-F in 25—125 gal. water per acre. AERIAL Spraying—Use MERTECT 340-F in 3—10 gal. per acre.

Begin application when disease first appears. Repeat application at 14—21 day intervals (up to a maximum of 5 applications). Do not apply within 21 days of harvest.

SUGAR BEETS (Post Harvest Treatment)—**Reduce rot caused by *Penicillium*, *Botrytis*, and *Fusarium* species:** Treat sugar beet roots as they enter storage with 0.42 fl. oz. of MERTECT 340-F to each 2,000 lb. of sugar beet roots in sufficient water for complete coverage. (0.42 fl. oz. of MERTECT 340-F is equivalent to 0.2 oz. of 2-(4-thiazolyl) benzimidazole.) Roots should be treated within 72 hours after lifting.

ORNAMENTAL BULBS AND CORMS—**Control Fusarium Basal Rot and Penicillium Blue Mold:** Clean and treat bulbs or corms within 24—48 hours of digging. Prepare suspension of 30 fl. oz. MERTECT 340-F per 100 gal. water. Submerge stock completely in dipping suspension. Suspension temperature should be 55—75°F (13—24°C). Discard suspension (1) when it becomes dirty, (2) after use five times, or (3) after 24 hours, whichever occurs first.

Fusarium (Bulbs and Corms): Dip bulbs 15—30 minutes; corms 15 minutes.

Blue Mold (Bulbs): Dip 10—15 minutes.

After treatment, dry bulbs or corms in a shaded, well ventilated area. Curing or retarding may precede or follow treatment.

SWEET POTATO SEED ROOTS—**Control Black Rot, Scurf, and Foot Rot:** Dip the seed roots in a suspension containing 8 fl. oz. MERTECT 340-F per 7.5 gal. water (107 fl. oz. per 100 gal.). Treat the seed roots for 1—2 minutes and plant immediately. Discard the suspension when the volume is too low or when it becomes dirty. Do not use the treated roots for food or feed.

POTATO—**Control Fusarium Tuber Rot:** Mist unwashed tubers on a conveyor line, with tumbling action, entering storage with 0.42 fl. oz. of MERTECT 340-F to each 2000 lb. of tubers in sufficient water for complete coverage. If an additional treatment is necessary before shipping and cutting the seed tubers, dip the tubers for 20 seconds, or mist at the same rate.

WHEAT—**Control of *Cercosporella* Foot Rot**—Apply MERTECT 340-F after the development of 2—3 tillers in the Fall but prior to the time the first node is visible in the Spring. AERIAL Application: Apply 16—24 fluid ounces of MERTECT 340-F in a minimum of 5 gallons water per acre. Use the high rate in fields with a previous history of foot rot. GROUND Application: Apply 16—24 fluid ounces of MERTECT 340-F per acre in sufficient water for coverage. Use the high rate in fields with a previous history of foot rot. Do not allow livestock to graze or feed on treated green wheat forage.

RICE—**Reduce Severity of Rice Blast, Stem Rot, Sheath Blight, and Narrow Brown Leaf Spot:** Apply MERTECT 340-F at 6.0—12.0 fl. oz. per acre per application in sufficient water for coverage (minimum of 5 gallons of water per acre for aerial application). Make two applications per season, the first application at boot initiation followed by a second application 14—21 days later. Use the higher rate when disease infestations are moderate to severe. Do not use in California.

CARROTS—**Control of Botrytis cinerla gray mold and Sclerotinia sclerotiorum (sclerotinia rot)**—Dip carrots, before storage, for 5—10 seconds in a suspension containing 42 fl. oz. of MERTECT 340-F per 100 gal. of water for each 2000 lb. of carrots.

The run-off and wastes from the dipping operation should not be discarded in a drainage which could contaminate public water systems.

BANANAS AND PLANTAINS—**Control of Crown Rot**—Use 45 ml (1.5 fl. oz.) to 100 ml (3.4 fl. oz.) per 227 liters (60 gallons) of water. Dip fruit for 2 to 4 minutes after it has been dehanded from the stem and passed through the delatexing operation. Use the low rate where rot incidence is low. Stir the mixture frequently to maintain the suspension.

STORAGE AND DISPOSAL

DISPOSAL: **Prohibitions**—Do not contaminate water, food, or feed by storage or disposal. Open dumping is prohibited. Do not reuse empty container. **Pesticide Disposal**—Pesticide, spray mixture, or rinsate that cannot be used or chemically reprocessed should be disposed of in a landfill approved for pesticides or buried in a safe place away from water supplies. **Container Disposal**—Triple rinse (or equivalent) and dispose in an incinerator or landfill approved for pesticide containers, or bury in a safe place. **General**—Consult Federal, state, or local disposal authorities for approved alternative procedures such as limited open burning.

NOTICE TO USER—Buyer assumes all risks of use and handling which are at variance in any way with the directions hereon. There are no warranties which extend beyond the description on this label.

DO NOT STORE BELOW 32°F (0°C)

SHAKE OR STIR WELL BEFORE USE

1-10-81

MSD AGVET
Division of Merck & Co., Inc.
Rahway, New Jersey 07065, U.S.A.

Lot

ACCEPTED
MAR 16 1982
Under the Federal Insecticide, Fungicide, and Rodenticide Act
For the pesticide described

Product
48015

Flowable

(Water Dispersible Suspension)

MERTECT® 340-F

FUNGICIDE

Active Ingredient	
2-(4-thiazolyl) benzimidazole	42.28%
Inert Ingredients	57.72%

EPA Est. 618-NJ-1

EPA REG No. 618-75-A

PRECAUTIONARY STATEMENTS

Hazards to Humans and Domestic Animals

KEEP OUT OF REACH OF CHILDREN

CAUTION

HARMFUL IF SWALLOWED

Do not get in eyes or on skin.

FIRST AID—**If in eyes or on skin:** Flush with water; for eyes, get medical attention.

Environmental Hazards

This pesticide is toxic to fish. Do not apply when weather conditions favor runoff or drift from the target area. Do not apply directly to lakes, streams or ponds. Do not contaminate water by cleaning of equipment or disposal of wastes.

See Other Panels for Additional Directions for Use

U.S. Pat. 3,370,957

2 GALLONS (7.57 liters)