3-21-94

MRID No. 429186-27

DATA EVALUATION RECORD

1. **CHEMICAL:** Fipronil and derivatives (M&B 46030). Shaughnessey No. 129121.

2. **TEST MATERIAL:** M&B 46030; Lot No. 78 GC90; 96.7% active ingredient; a white powder.

3. **STUDY TYPE:** 72-4. Freshwater Fish Early Life-Stage Test. Species Tested: Rainbow Trout (*Oncorhynchus mykiss*).

4. **CITATION:** Machado, M.W. 1992. (M&B 46030) - The Toxicity to Rainbow Trout (*Oncorhynchus mykiss*) During an Early Life-Stage Exposure. SLI Report No. 92-1-4084. Performed by Springborn Laboratories, Inc., Wareham, MA. Submitted by Rhone-Poulenc Ag Company, Research Triangle Park, NC. EPA MRID No. 429186-27.

5. **REVIEWED BY:**

Rosemary Graham Mora, M.S.
Associate Scientist
KBN Engineering and
Applied Sciences, Inc.

Signature: Rosemary Graham Mora
Date: 2/2/94

3/9/94

6. **APPROVED BY:**

Pim Kosalwat, Ph.D.
Senior Scientist
KBN Engineering and
Applied Sciences, Inc.

Signature: P. Kosalwat
Date: 2/4/94

James J. Goodyear, Ph.D.
Project Officer, EEB/EFED
USEPA

Signature: Goodyear
Date: 3 21 94

3/10/94

7. **CONCLUSIONS:** This study is scientifically sound and meets the guideline requirements for a fish early life-stage test. Based on the effects of M&B 46030 on larval length, the MATC for rainbow trout was >6.6 and <15 µg ai/l (geometric mean MATC = 9.9 µg ai/l).

8. **RECOMMENDATIONS:** N/A.

9. **BACKGROUND:**

10. **DISCUSSION OF INDIVIDUAL TESTS:** N/A.

11. MATERIALS AND METHODS:

A. Test Animals: Unfertilized rainbow trout (*Oncorhynchus mykiss*) eggs and sperm were obtained (separately packaged at 9°C; warmed to 11°C upon receipt) from Mount Lassen Trout Farm, Red Bluff, CA. The eggs were fertilized in a stainless steel bowl by adding the eggs to half of the sperm. The bowl was swirled to thoroughly mix the contents. The remaining sperm and a small amount of control water (11°C) were added to the bowl and swirled again. The contents were allowed to stand undisturbed for 2 minutes to complete fertilization. The excess sperm was rinsed from the eggs with control water. The eggs were allowed to harden for 1 hour. During the period of egg distribution, the temperature was allowed to rise to 12°C.

B. Test System: The test system was an intermittent-flow proportional diluter with a 50% dilution factor. The diluter delivered approximately 2 l of solution per cycle to each aquarium at an average rate of approximately 6.6 volume replacements per day.

Each glass test aquarium (39 x 20 x 25 cm) was equipped with a 19.5-cm high side drain that maintained a constant solution volume of 15 l. Embryo incubation cups were glass jars (5 cm O.D., 8 cm high) with 2000 micron-mesh Nitex® screen bottoms. A rocker arm apparatus was used to gently oscillate incubation cups in the test chambers.

The aquaria were impartially positioned in a water bath designed to maintain the solution temperature at 12 ±1°C. Sixteen hours of light at an intensity of 30-50 footcandles were provided each day. Sudden transitions from light to dark and dark to light were avoided.

The dilution water was well water which was pumped into an epoxy-coated concrete reservoir where it was supplemented with untreated Town of Wareham well water and aerated. The well water, characterized weekly, had total hardness and alkalinity ranges of 24-32 and 18-25 mg/l as $CaCO_3$, respectively, a pH range of 6.8-7.4, and a specific conductivity range of 100-150 µmhos/cm.

A stock solution (15.4 mg ai/ml) was prepared by dissolving 0.796 g (0.770 g ai) of test material in acetone to a final volume of 50 ml.

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C. **Dosage:** Ninety-day, flow-through test. Based on results of preliminary testing, nominal test concentrations selected were 6.2, 12, 25, 50, and 100 µg ai/l. A dilution water control and a solvent control (6.5 µl acetone/l) were also included. The concentration of acetone in the solvent control was equivalent to the highest solvent concentration in any exposure concentration.

D. **Design:** Fifty embryos were impartially distributed to each of 28 incubation cups. Two cups were suspended in each duplicate test chamber per treatment.

Embryo viability was determined on day 18, when all the eggs exhibited well-pronounced embryonic development. Due to the extreme sensitivity of the embryos to physical trauma, dead embryos were not removed from the incubation cups until test day 16. On day 18, ten live embryos were impartially selected from each of the two incubation cups (20 per replicate aquarium) and placed into one incubation cup. The remaining embryos were placed in the second cup of each replicate. On test day 30 (considered hatch day), the surviving larvae from the first incubation cup of each replicate were released into their respective aquarium. Unhatched embryos remained in the incubation cups until day 31. The surviving larvae from the second cup of each replicate were counted and discarded. Survival of organisms at hatch (number of live larvae/number of viable embryos) was determined on day 31 when hatching was considered complete.

When larvae reached swim-up stage and began actively feeding (day 13 post-hatch), they were fed live *Artemia* nauplii two to three times daily. The aquaria were brushed and siphoned when necessary.

Behavior and appearance of larvae were observed and recorded daily. Larval survival was determined twice weekly. At test termination (60 days post-hatch), the larvae were counted, and individually weighed and measured.

Dissolved oxygen concentration (DO), pH, and temperature for each replicate were measured daily. Temperature was continuously monitored in one replicate of the control. Conductivity, total hardness, and alkalinity were measured on day 0 and weekly thereafter in alternating replicates of the highest and lowest exposure groups and the control.

Water samples were collected from the midpoint of each aquarium on test day 0 and at least weekly thereafter for determination of M&B 46030 concentrations. Samples were analyzed using high performance liquid chromatography.

E. **Statistics:** The percentage survival data were transformed (arcsine square-root percentage) before analysis. All statistical analyses were performed using the mean organism response in each replicate aquarium. All data were tested for assumptions of homogeneity of variance (Bartlett's test). A one-way, single-classification analysis of variance (ANOVA) was used to determine significant differences between the dilution water control and the solvent control. No significant differences were determined, therefore, the control data were pooled for comparison to the exposure concentrations. Williams' test was used to assess treatment effects. Treatment levels that caused significant survival effects were excluded from the analysis of growth data. All statistical conclusions were made at 95% confidence level, except for Bartlett's test which was done at 99% level.

12. **REPORTED RESULTS:** Mean measured concentrations were 2.6, 6.6, 15, 26, and 60 µg ai/l which represent 42-60% of nominal concentrations (Tables 2 and 3, attached).

No significant difference in embryo viability or hatchability was observed between the exposure concentrations and the pooled control (Table 5, attached). Survival at 26 and 60 µg ai/l was significantly reduced when compared to that of the pooled control.

Larval length at 2.6 and 15 µg ai/l was significantly reduced when compared to that of the pooled control. However, the reduction at both levels was considered biologically insignificant. "The absence of a concentration-response relationship, for organism length, at these lower treatment levels (i.e., 2.6-15 µg ai/l) and no statistical difference between larval weight at the same exposure concentrations, corroborates the lack of biological significance between the growth of rainbow trout exposed to ≤15 µg ai/l M&B 46030 and the control solutions." Wet weight at concentration ≤15 µg ai/l was not significantly reduced when compared to pooled control data.

During the study, the test solutions had a pH range of 6.6-7.4, a mean DO of 9.6-10.0 mg/l, and a mean total hardness and alkalinity of 30-31 and 27-28 mg/l as $CaCO_3$,

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respectively. Minimum/maximum temperature readings ranged from 10 to 19°C.

13. **STUDY AUTHOR'S CONCLUSIONS/QUALITY ASSURANCE MEASURES:** "Based on the statistically significant ($p \leq 0.05$) reduction in larval survival following 60 days post-hatch exposure, the Lowest Observed Effect Concentration (LOEC) was determined to be 26 µg ai/l and the No Observed Effect Concentration (NOEC) was determined to be 15 µg ai/l. Based on these results, the Maximum Acceptable Toxicant Concentration (MATC) for M&B 46030 and rainbow trout embryos and larvae is estimated to be ≥15 and ≤26 µg ai/l (geometric mean MATC = 20 µg ai/l)."

Good laboratory practice (GLP) compliance and quality assurance statements were included in the report indicating that the study was performed in accordance with EPA GLP regulations (40 CFR Part 160), except in the case of stability, characterization, and verification of test substance identity.

14. **REVIEWER'S DISCUSSION AND INTERPRETATION OF STUDY RESULTS:**

A. **Test Procedure:** The test procedure is generally in accordance with the SEP and ASTM guidelines, except for the following deviations:

Raw water quality data were not included in the report as required.

Only two replicates per treatment level were used during the study; the SEP requires four replicates.

The total hardness (24-32 mg/l as $CaCO_3$) of the dilution water was slightly lower than the recommended hardness (40-48 mg/l as $CaCO_3$).

The light intensity employed in this study was 323-538 lux at the water surface. The SEP recommends the intensity of 400-800 lux.

The continuous test temperature ranged from 10 to 19°C; the SEP recommends 10 ±2°C during the exposure period.

Embryos were "impartially" distributed; the SEP requires random distribution of test organisms to the test vessels.

B. **Statistical Analysis:** Survival data were analyzed using Toxstat® (Version 3.3). The data failed the

assumptions of homogeneity of variance (Bartletts or Hartley tests) or normality (Chi-square or Shapiro Wilks tests), therefore a non-parametric test (Kruskal-Wallis) was used to evaluate treatment effects. The reviewer's analysis of survival data showed no significant difference in exposure concentrations when compared to the control (printout attached). However, upon visual examination of the data, it is obvious that there is a substantial reduction in survival (78 and 0% survival) at the two highest concentrations when compared to the control (98% survival).

Individual length and weight data were analyzed using a 2-way analysis of variance (ANOVA) coupled with Bonferroni's test and a contrast test (Systat®) for treatment comparisons (printout, attached). The results of Bonferroni's test for weight and length data showed a significant reduction at 26 µg ai/l when compared to the controls. The results of the contrast comparison tests showed a significant reduction in length at 2.6, 15, and 26 µg ai/l and a significant reduction in weight at 26 µg ai/l when compared to the dilution water control.

C. **Discussion/Results:** Although the temperature of the test solutions ranged from 10 to 19°C, it is not likely that this deviation had a significant effect on the growth of the test organisms. This temperature range (10-19°C) represents two excursions from the general temperature (10-14°C) which each lasted 12 hours or less and were caused by a malfunction of the chiller of the water bath.

The author's statistical analysis of length data demonstrated a significant reduction at 2.6 and 15.0 µg ai/l from the pooled control, a difference which the author claimed to be biologically insignificant. The reviewer's results showed a significant reduction in length at 2.6, 15.0, and 26.0 µg ai/l. The reviewer agrees with the author that the difference at 2.6 µg ai/l is probably not due to the treatment since length at 6.6 µg ai/l was not statistically reduced. However, the reduction at 15 µg ai/l may be treatment related, therefore, a conservative estimate of the NOEC would be 6.6 µg ai/l.

This study is scientifically sound and meets the guideline requirements for a fish early life-stage test. Based on the effects of M&B 46030 on larval

length, the MATC for rainbow trout was >6.6 and <15 µg ai/l (geometric mean MATC = 9.9 µg ai/l).

D. **Adequacy of the Study:**

(1) Classification: Core.

(2) Rationale: N/A.

(3) Repairability: N/A.

15. **COMPLETION OF ONE-LINER:** Yes; 14 January 1994.

M&B 46030: Survival of Exposed P.promelas Larvae
File: 42918627.sur Transform: ARC SINE(SQUARE ROOT(Y))

Chi-square test for normality: actual and expected frequencies

INTERVAL	<-1.5	-1.5 to <-0.5	-0.5 to 0.5	>0.5 to 1.5	>1.5
EXPECTED	0.938	3.388	5.348	3.388	0.938
OBSERVED	0	6	2	6	0

Calculated Chi-Square goodness of fit test statistic = 7.9994
Table Chi-Square value (alpha = 0.01) = 13.277

Data PASS normality test. Continue analysis.

M&B 46030: Survival of Exposed P.promelas Larvae
File: 42918627.sur Transform: ARC SINE(SQUARE ROOT(Y))

Shapiro Wilks test for normality

D = 0.032

W = 0.814

Critical W (P = 0.05) (n = 14) = 0.874
Critical W (P = 0.01) (n = 14) = 0.825

Data FAIL normality test. Try another transformation.

Warning - The two homogeneity tests are sensitive to non-normal data and should not be performed.

M&B 46030: Survival of Exposed P.promelas Larvae
: 42918627.sur Transform: ARC SINE(SQUARE ROOT(Y))

Hartley test for homogeneity of variance
Bartletts test for homogeneity of variance

These two tests can not be performed because at least one group has zero variance.

Data FAIL to meet homogeneity of variance assumption.
Additional transformations are useless.

TITLE: M&B 46030: Survival of Exposed P.promelas Larvae
FILE: 42918627.sur
TRANSFORM: ARC SINE(SQUARE ROOT(Y)) NUMBER OF GROUPS: 7

GRP	IDENTIFICATION	REP	VALUE	TRANS VALUE
1	Control	1	1.0000	1.4588
1	Control	2	0.9500	1.3453
2	Solvent Control	1	1.0000	1.4588
2	Solvent Control	2	0.9500	1.3453
3	2.6 ug ai/l	1	1.0000	1.4588
3	2.6 ug ai/l	2	0.9500	1.3453
4	6.6 ug ai/l	1	1.0000	1.4588
4	6.6 ug ai/l	2	0.9500	1.3453
5	15 ug ai/l	1	0.9500	1.3453
5	15 ug ai/l	2	0.9000	1.2490
6	26 ug ai/l	1	0.8000	1.1071
6	26 ug ai/l	2	0.7500	1.0472
7	60 ug ai/l	1	0.0000	0.1120
7	60 ug ai/l	2	0.0000	0.1120

M&B 46030: Survival of Exposed P.promelas Larvae
File: 42918627.sur Transform: ARC SINE(SQUARE ROOT(Y))

KRUSKAL-WALLIS ANOVA BY RANKS - TABLE 1 OF 2 (p=0.05)

GROUP	IDENTIFICATION	TRANSFORMED MEAN	MEAN CALCULATED IN ORIGINAL UNITS	RANK SUM
1	Control	1.402	0.975	20.500
2	Solvent Control	1.402	0.975	20.500
3	2.6 ug ai/l	1.402	0.975	20.500
4	6.6 ug ai/l	1.402	0.975	20.500
5	15 ug ai/l	1.297	0.925	13.000
6	26 ug ai/l	1.077	0.775	7.000
	60 ug ai/l	0.112	0.000	3.000

Calculated H Value = 10.210 Critical H Value Table = 12.590
Since Calc H < Crit H FAIL TO REJECT Ho:All groups are equal.

M&B 46030: Survival of Exposed P.promelas Larvae
File: 42918627.sur Transform: ARC SINE(SQUARE ROOT(Y))

DUNNS MULTIPLE COMPARISON - KRUSKAL-WALLIS - TABLE 2 OF 2 (p=0.05)

GROUP	IDENTIFICATION	TRANSFORMED MEAN	ORIGINAL MEAN	GROUP 0 0 0 0 0 0 0 7 6 5 4 1 2 3
7	60 ug ai/l	0.112	0.000	\
6	26 ug ai/l	1.077	0.775	. \
5	15 ug ai/l	1.297	0.925	. . \
4	6.6 ug ai/l	1.402	0.975	. . . \
1	Control	1.402	0.975	. . . . \
2	Solvent Control	1.402	0.975	. . . . . \
3	2.6 ug ai/l	1.402	0.975	. . . . . \

* = significant difference (p=0.05) . = no significant difference
Table q value (0.05,7) = 3.038 SE = 4.038

TRT 1 = ~~Solvent~~ Dilution Water Control

TRT 2 = ~~Dilution Water~~ Solvent Control

TRT 3 = 6.2 ug ai/l nominal

TRT 4 = 12 ug ai/l "

TRT 5 = 25 ug ai/l "

TRT 6 = 50 ug ai/l "

		TRT	REP	LENGTH	WEIGHT
CASE	1	1.0000	1.0000	59.0000	2.0184
CASE	2	1.0000	1.0000	68.0000	2.9042
CASE	3	1.0000	1.0000	67.0000	2.9512
CASE	4	1.0000	1.0000	62.0000	2.1924
CASE	5	1.0000	1.0000	63.0000	2.4922
CASE	6	1.0000	1.0000	60.0000	1.8659
CASE	7	1.0000	1.0000	59.0000	1.8044
CASE	8	1.0000	1.0000	65.0000	2.8730
CASE	9	1.0000	1.0000	68.0000	3.1342
CASE	10	1.0000	1.0000	58.0000	2.0128
CASE	11	1.0000	1.0000	61.0000	2.1073
CASE	12	1.0000	1.0000	52.0000	1.5091
CASE	13	1.0000	1.0000	67.0000	2.6213
CASE	14	1.0000	1.0000	61.0000	2.1185
CASE	15	1.0000	1.0000	66.0000	2.4689
CASE	16	1.0000	1.0000	65.0000	2.4956
CASE	17	1.0000	1.0000	58.0000	1.7375
CASE	18	1.0000	1.0000	58.0000	1.7073
CASE	19	1.0000	1.0000	58.0000	2.1178
CASE	20	1.0000	2.0000	61.0000	2.3766
CASE	21	1.0000	2.0000	58.0000	2.0160
CASE	22	1.0000	2.0000	63.0000	2.5023
CASE	23	1.0000	2.0000	61.0000	2.3050
CASE	24	1.0000	2.0000	59.0000	2.0099
CASE	25	1.0000	2.0000	61.0000	2.4929
CASE	26	1.0000	2.0000	57.0000	1.7915
CASE	27	1.0000	2.0000	63.0000	2.3305
CASE	28	1.0000	2.0000	61.0000	2.2203
CASE	29	1.0000	2.0000	58.0000	1.9859
CASE	30	1.0000	2.0000	57.0000	1.9055
CASE	31	1.0000	2.0000	39.0000	0.4772
CASE	32	1.0000	2.0000	63.0000	2.4049
CASE	33	1.0000	2.0000	64.0000	2.3151
CASE	34	1.0000	2.0000	58.0000	1.9241
CASE	35	1.0000	2.0000	64.0000	2.5178
CASE	36	1.0000	2.0000	59.0000	1.9004
CASE	37	1.0000	2.0000	72.0000	3.7679
CASE	38	1.0000	2.0000	59.0000	2.1221
CASE	39	1.0000	2.0000	58.0000	2.0005
CASE	40	2.0000	1.0000	62.0000	2.4568
CASE	41	2.0000	1.0000	63.0000	2.2853
CASE	42	2.0000	1.0000	64.0000	2.6258
CASE	43	2.0000	1.0000	56.0000	1.6325
CASE	44	2.0000	1.0000	59.0000	2.1117
CASE	45	2.0000	1.0000	59.0000	1.9496
CASE	46	2.0000	1.0000	57.0000	2.1573
CASE	47	2.0000	1.0000	64.0000	2.3289
CASE	48	2.0000	1.0000	61.0000	1.9198
CASE	49	2.0000	1.0000	59.0000	1.9873
CASE	50	2.0000	1.0000	62.0000	2.2965
CASE	51	2.0000	1.0000	59.0000	1.9357
CASE	52	2.0000	1.0000	60.0000	2.0329

CASE	53	2.0000	1.0000	59.0000	2.1616
CASE	54	2.0000	1.0000	58.0000	1.8451
CASE	55	2.0000	1.0000	59.0000	2.1197
CASE	56	2.0000	1.0000	58.0000	1.7856
CASE	57	2.0000	1.0000	63.0000	2.4197
CASE	58	2.0000	1.0000	61.0000	1.9661
CASE	59	2.0000	2.0000	61.0000	2.5143
CASE	60	2.0000	2.0000	62.0000	2.2648
CASE	61	2.0000	2.0000	60.0000	2.1687
CASE	62	2.0000	2.0000	59.0000	2.0143
CASE	63	2.0000	2.0000	58.0000	1.9720
CASE	64	2.0000	2.0000	59.0000	1.9948
CASE	65	2.0000	2.0000	60.0000	2.3425
CASE	66	2.0000	2.0000	53.0000	1.4280
CASE	67	2.0000	2.0000	56.0000	1.6978
CASE	68	2.0000	2.0000	62.0000	2.3799
CASE	69	2.0000	2.0000	63.0000	2.4058
CASE	70	2.0000	2.0000	61.0000	2.0680
CASE	71	2.0000	2.0000	62.0000	2.2360
CASE	72	2.0000	2.0000	59.0000	2.2153
CASE	73	2.0000	2.0000	58.0000	1.9963
CASE	74	2.0000	2.0000	63.0000	2.4272
CASE	75	2.0000	2.0000	56.0000	1.6888
CASE	76	2.0000	2.0000	60.0000	1.9401
CASE	77	2.0000	2.0000	64.0000	2.4028
CASE	78	2.0000	2.0000	56.0000	1.7299
CASE	79	3.0000	1.0000	54.0000	2.1309
CASE	80	3.0000	1.0000	62.0000	2.2239
CASE	81	3.0000	1.0000	65.0000	2.8293
CASE	82	3.0000	1.0000	50.0000	1.5195
CASE	83	3.0000	1.0000	51.0000	2.0544
CASE	84	3.0000	1.0000	60.0000	2.1598
CASE	85	3.0000	1.0000	54.0000	2.2389
CASE	86	3.0000	1.0000	65.0000	2.5677
CASE	87	3.0000	1.0000	60.0000	2.1518
CASE	88	3.0000	1.0000	52.0000	1.8591
CASE	89	3.0000	1.0000	54.0000	1.4865
CASE	90	3.0000	1.0000	64.0000	2.4456
CASE	91	3.0000	1.0000	59.0000	2.1462
CASE	92	3.0000	1.0000	52.0000	1.3032
CASE	93	3.0000	1.0000	63.0000	2.7105
CASE	94	3.0000	1.0000	63.0000	2.2636
CASE	95	3.0000	1.0000	51.0000	1.7073
CASE	96	3.0000	1.0000	65.0000	2.7045
CASE	97	3.0000	1.0000	60.0000	2.2563
CASE	98	3.0000	1.0000	53.0000	1.9363
CASE	99	3.0000	2.0000	57.0000	1.7200
CASE	100	3.0000	2.0000	55.0000	2.3392
CASE	101	3.0000	2.0000	62.0000	2.3467
CASE	102	3.0000	2.0000	58.0000	2.1312
CASE	103	3.0000	2.0000	59.0000	2.0862
CASE	104	3.0000	2.0000	58.0000	1.9607
CASE	105	3.0000	2.0000	65.0000	2.5621
CASE	106	3.0000	2.0000	63.0000	2.4596
CASE	107	3.0000	2.0000	60.0000	1.9556
CASE	108	3.0000	2.0000	57.0000	2.0618
CASE	109	3.0000	2.0000	56.0000	2.2787
CASE	110	3.0000	2.0000	60.0000	2.0037
CASE	111	3.0000	2.0000	59.0000	2.1247
CASE	112	3.0000	2.0000	52.0000	1.8789
CASE	113	3.0000	2.0000	53.0000	1.7492
CASE	114	3.0000	2.0000	64.0000	2.5381
CASE	115	3.0000	2.0000	56.0000	2.1130
CASE	116	3.0000	2.0000	58.0000	2.0488
CASE	117	3.0000	2.0000	59.0000	2.2896
CASE	118	4.0000	1.0000	65.0000	2.8410

CASE	119	4.0000	1.0000	63.0000	2.4224
CASE	120	4.0000	1.0000	59.0000	1.8317
CASE	121	4.0000	1.0000	60.0000	2.0447
CASE	122	4.0000	1.0000	62.0000	2.3103
CASE	123	4.0000	1.0000	62.0000	2.0585
CASE	124	4.0000	1.0000	59.0000	2.0887
CASE	125	4.0000	1.0000	57.0000	1.8493
CASE	126	4.0000	1.0000	59.0000	2.0620
CASE	127	4.0000	1.0000	57.0000	1.9968
CASE	128	4.0000	1.0000	58.0000	1.8490
CASE	129	4.0000	1.0000	59.0000	2.1758
CASE	130	4.0000	1.0000	58.0000	1.5330
CASE	131	4.0000	1.0000	60.0000	2.0160
CASE	132	4.0000	1.0000	60.0000	2.1176
CASE	133	4.0000	1.0000	61.0000	2.0980
CASE	134	4.0000	1.0000	63.0000	2.7727
CASE	135	4.0000	1.0000	56.0000	1.7871
CASE	136	4.0000	1.0000	60.0000	2.2197
CASE	137	4.0000	1.0000	58.0000	1.9759
CASE	138	4.0000	2.0000	65.0000	2.8434
CASE	139	4.0000	2.0000	61.0000	2.1875
CASE	140	4.0000	2.0000	55.0000	1.9014
CASE	141	4.0000	2.0000	58.0000	2.1235
CASE	142	4.0000	2.0000	64.0000	2.4352
CASE	143	4.0000	2.0000	63.0000	2.7455
CASE	144	4.0000	2.0000	61.0000	2.1321
CASE	145	4.0000	2.0000	58.0000	1.9478
CASE	146	4.0000	2.0000	60.0000	2.0346
CASE	147	4.0000	2.0000	63.0000	2.3820
CASE	148	4.0000	2.0000	56.0000	1.7395
CASE	149	4.0000	2.0000	58.0000	1.9617
CASE	150	4.0000	2.0000	59.0000	2.0867
CASE	151	4.0000	2.0000	57.0000	1.9550
CASE	152	4.0000	2.0000	58.0000	1.9062
CASE	153	4.0000	2.0000	62.0000	2.3528
CASE	154	4.0000	2.0000	61.0000	2.2753
CASE	155	4.0000	2.0000	62.0000	2.7297
CASE	156	4.0000	2.0000	58.0000	1.9854
CASE	157	5.0000	1.0000	60.0000	1.9224
CASE	158	5.0000	1.0000	62.0000	2.4270
CASE	159	5.0000	1.0000	61.0000	2.2892
CASE	160	5.0000	1.0000	59.0000	2.3693
CASE	161	5.0000	1.0000	65.0000	3.0089
CASE	162	5.0000	1.0000	60.0000	2.3342
CASE	163	5.0000	1.0000	61.0000	2.5554
CASE	164	5.0000	1.0000	50.0000	1.3835
CASE	165	5.0000	1.0000	56.0000	2.0277
CASE	166	5.0000	1.0000	57.0000	1.9054
CASE	167	5.0000	1.0000	59.0000	1.8905
CASE	168	5.0000	1.0000	56.0000	1.6257
CASE	169	5.0000	1.0000	59.0000	1.9689
CASE	170	5.0000	1.0000	58.0000	2.1062
CASE	171	5.0000	1.0000	54.0000	1.5689
CASE	172	5.0000	1.0000	59.0000	1.9713
CASE	173	5.0000	1.0000	63.0000	2.4546
CASE	174	5.0000	1.0000	56.0000	1.9098
CASE	175	5.0000	1.0000	55.0000	1.7398
CASE	176	5.0000	2.0000	59.0000	2.2797
CASE	177	5.0000	2.0000	58.0000	2.0629
CASE	178	5.0000	2.0000	67.0000	3.1215
CASE	179	5.0000	2.0000	59.0000	2.0080
CASE	180	5.0000	2.0000	60.0000	2.0834
CASE	181	5.0000	2.0000	57.0000	2.0005
CASE	182	5.0000	2.0000	60.0000	2.3947
CASE	183	5.0000	2.0000	57.0000	1.8599
CASE	184	5.0000	2.0000	58.0000	2.1013

12

CASE	185	5.0000	2.0000	54.0000	1.8913
CASE	186	5.0000	2.0000	50.0000	1.1821
CASE	187	5.0000	2.0000	58.0000	1.9492
CASE	188	5.0000	2.0000	56.0000	1.8872
CASE	189	5.0000	2.0000	61.0000	2.3735
CASE	190	5.0000	2.0000	54.0000	1.8578
CASE	191	5.0000	2.0000	60.0000	2.3265
CASE	192	5.0000	2.0000	61.0000	2.4760
CASE	193	5.0000	2.0000	57.0000	1.8513
CASE	194	6.0000	1.0000	27.0000	0.5724
CASE	195	6.0000	1.0000	60.0000	2.4954
CASE	196	6.0000	1.0000	57.0000	2.0504
CASE	197	6.0000	1.0000	37.0000	1.1052
CASE	198	6.0000	1.0000	38.0000	0.9652
CASE	199	6.0000	1.0000	52.0000	1.8383
CASE	200	6.0000	1.0000	60.0000	2.2801
CASE	201	6.0000	1.0000	57.0000	2.4415
CASE	202	6.0000	1.0000	58.0000	2.0557
CASE	203	6.0000	1.0000	51.0000	1.5620
CASE	204	6.0000	1.0000	56.0000	1.9092
CASE	205	6.0000	1.0000	51.0000	1.6476
CASE	206	6.0000	1.0000	41.0000	0.9155
CASE	207	6.0000	1.0000	57.0000	1.8582
CASE	208	6.0000	1.0000	36.0000	0.5597
CASE	209	6.0000	2.0000	55.0000	1.9588
CASE	210	6.0000	2.0000	53.0000	1.6930
CASE	211	6.0000	2.0000	60.0000	2.6144
CASE	212	6.0000	2.0000	41.0000	1.3414
CASE	213	6.0000	2.0000	50.0000	1.8974
CASE	214	6.0000	2.0000	58.0000	2.5183
CASE	215	6.0000	2.0000	56.0000	1.7978
CASE	216	6.0000	2.0000	58.0000	1.9550
CASE	217	6.0000	2.0000	41.0000	1.1683
CASE	218	6.0000	2.0000	53.0000	1.7232
CASE	219	6.0000	2.0000	55.0000	1.8575
CASE	220	6.0000	2.0000	47.0000	1.5572
CASE	221	6.0000	2.0000	32.0000	0.5257
CASE	222	6.0000	2.0000	46.0000	1.5460
CASE	223	6.0000	2.0000	50.0000	1.4117

ANOVA on Weights

LEVELS ENCOUNTERED DURING PROCESSING ARE:
TRT
1.0000 2.0000 3.0000 4.0000 5.0000 6.0000
REP
1.0000 2.0000

DEP VAR: WEIGHT N: 223 MULTIPLE R: 0.393 SQUARED MULTIPLE R: 0.155

ANALYSIS OF VARIANCE

SOURCE	SUM-OF-SQUARES	DF	MEAN-SQUARE	F-RATIO	P
TRT	6.3534	5	1.2707	7.4604	0.0000
REP	0.0124	1	0.0124	0.0727	0.7877
TRT*REP	0.2399	5	0.0480	0.2817	0.9228
ERROR	35.9381	211	0.1703		

DURBIN-WATSON D STATISTIC 2.071
FIRST ORDER AUTOCORRELATION -.037

Post-hoc pairwise comparison of weight/Bonferroni.
USING LEAST SQUARES MEANS.
POST HOC TEST OF WEIGHT

MATRIX OF PAIRWISE MEAN DIFFERENCES:

	1	2	3	4	5
1	0.0000				
2	-0.1189	0.0000			
3	-0.0822	0.0368	0.0000		
4	-0.0699	0.0490	0.0122	0.0000	
5	-0.1334	-0.0145	-0.0513	-0.0635	0.0000
6	-0.5585	-0.4395	-0.4763	-0.4886	-0.4251

	6
6	0.0000

BONFERRONI ADJUSTMENT.
MATRIX OF PAIRWISE COMPARISON PROBABILITIES:

	1	2	3	4	5
1	1.0000				
2	1.0000	1.0000			
3	1.0000	1.0000	1.0000		
4	1.0000	1.0000	1.0000	1.0000	
5	1.0000	1.0000	1.0000	1.0000	1.0000
6	0.0000	0.0003	0.0001	0.0000	0.0006

	6
6	1.0000

ANOVA on Lengths

LEVELS ENCOUNTERED DURING PROCESSING ARE:
TRT
1.0000 2.0000 3.0000 4.0000 5.0000 6.0000
REP
1.0000 2.0000

DEP VAR: LENGTH N: 223 MULTIPLE R: 0.584 SQUARED MULTIPLE R: 0.342

ANALYSIS OF VARIANCE

SOURCE	SUM-OF-SQUARES	DF	MEAN-SQUARE	F-RATIO	P
TRT	2613.7275	5	522.7455	21.4541	0.0000
REP	1.7088	1	1.7088	0.0701	0.7914
TRT*REP	57.3201	5	11.4640	0.4705	0.7980
ERROR	5141.1795	211	24.3658		

DURBIN-WATSON D STATISTIC 2.052
FIRST ORDER AUTOCORRELATION -.027

Post-hoc pairwise comparison of length/Bonferroni.
USING LEAST SQUARES MEANS.
POST HOC TEST OF LENGTH

MATRIX OF PAIRWISE MEAN DIFFERENCES:

	1	2	3	4	5
1	0.0000				
2	-0.9171	0.0000			
3	-2.6342	-1.7171	0.0000		
4	-0.9224	-0.0053	1.7118	0.0000	
5	-2.5300	-1.6129	0.1042	-1.6076	0.0000
6	-11.0294	-10.1123	-8.3952	-10.1070	-8.4994

	6
6	0.0000

BONFERRONI ADJUSTMENT.
MATRIX OF PAIRWISE COMPARISON PROBABILITIES:

	1	2	3	4	5
1	1.0000				
2	1.0000	1.0000			
3	0.2910	1.0000	1.0000		
4	1.0000	1.0000	1.0000	1.0000	
5	0.3994	1.0000	1.0000	1.0000	1.0000
6	0.0000	0.0000	0.0000	0.0000	0.0000

	6
6	1.0000

THE FOLLOWING RESULTS ARE FOR:
TRT = 1.0000
REP = 1.0000

TOTAL OBSERVATIONS: 19

	WEIGHT	LENGTH
N OF CASES	19	19
MINIMUM	1.5091	52.0000
MAXIMUM	3.1342	68.0000
MEAN	2.2701	61.8421
STANDARD DEV	0.4718	4.3750

THE FOLLOWING RESULTS ARE FOR:
TRT = 1.0000
REP = 2.0000

TOTAL OBSERVATIONS: 20

	WEIGHT	LENGTH
N OF CASES	20	20
MINIMUM	0.4772	39.0000
MAXIMUM	3.7679	72.0000
MEAN	2.1683	59.7500
STANDARD DEV	0.5795	5.9989

THE FOLLOWING RESULTS ARE FOR:
TRT = 2.0000
REP = 1.0000

TOTAL OBSERVATIONS: 19

	WEIGHT	LENGTH
N OF CASES	19	19
MINIMUM	1.6325	56.0000
MAXIMUM	2.6258	64.0000
MEAN	2.1062	60.1579
STANDARD DEV	0.2514	2.3396

THE FOLLOWING RESULTS ARE FOR:
TRT = 2.0000
REP = 2.0000

TOTAL OBSERVATIONS: 20

	WEIGHT	LENGTH
N OF CASES	20	20
MINIMUM	1.4280	53.0000
MAXIMUM	2.5143	64.0000
MEAN	2.0944	59.6000
STANDARD DEV	0.2952	2.8359

THE FOLLOWING RESULTS ARE FOR:
TRT = 3.0000
REP = 1.0000

TOTAL OBSERVATIONS: 20

	WEIGHT	LENGTH
N OF CASES	20	20
MINIMUM	1.3032	50.0000
MAXIMUM	2.8293	65.0000
MEAN	2.1348	57.8500
STANDARD DEV	0.4148	5.4703

THE FOLLOWING RESULTS ARE FOR:
TRT = 3.0000
REP = 2.0000

TOTAL OBSERVATIONS: 19

	WEIGHT	LENGTH
N OF CASES	19	19
MINIMUM	1.7200	52.0000
MAXIMUM	2.5621	65.0000
MEAN	2.1394	58.4737
STANDARD DEV	0.2430	3.4378

THE FOLLOWING RESULTS ARE FOR:
TRT = 4.0000
REP = 1.0000

TOTAL OBSERVATIONS: 20

	WEIGHT	LENGTH
N OF CASES	20	20
MINIMUM	1.5330	56.0000
MAXIMUM	2.8410	65.0000
MEAN	2.1025	59.8000
STANDARD DEV	0.3101	2.3079

THE FOLLOWING RESULTS ARE FOR:
TRT = 4.0000
REP = 2.0000

TOTAL OBSERVATIONS: 19

	WEIGHT	LENGTH
N OF CASES	19	19
MINIMUM	1.7395	55.0000
MAXIMUM	2.8434	65.0000
MEAN	2.1961	59.9474
STANDARD DEV	0.3139	2.7983

THE FOLLOWING RESULTS ARE FOR:
TRT = 5.0000
REP = 1.0000

TOTAL OBSERVATIONS: 19

	WEIGHT	LENGTH
N OF CASES	19	19
MINIMUM	1.3835	50.0000
MAXIMUM	3.0089	65.0000
MEAN	2.0768	58.4211
STANDARD DEV	0.3914	3.4851

THE FOLLOWING RESULTS ARE FOR:
TRT = 5.0000
REP = 2.0000

TOTAL OBSERVATIONS: 18

	WEIGHT	LENGTH
N OF CASES	18	18
MINIMUM	1.1821	50.0000
MAXIMUM	3.1215	67.0000
MEAN	2.0948	58.1111
STANDARD DEV	0.3897	3.5627

THE FOLLOWING RESULTS ARE FOR:
TRT = 6.0000
REP = 1.0000

TOTAL OBSERVATIONS: 15

	WEIGHT	LENGTH
N OF CASES	15	15
MINIMUM	0.5597	27.0000
MAXIMUM	2.4954	60.0000
MEAN	1.6171	49.2000
STANDARD DEV	0.6467	10.5641

THE FOLLOWING RESULTS ARE FOR:
TRT = 6.0000
REP = 2.0000

TOTAL OBSERVATIONS: 15

	WEIGHT	LENGTH
N OF CASES	15	15
MINIMUM	0.5257	32.0000
MAXIMUM	2.6144	60.0000
MEAN	1.7044	50.3333
STANDARD DEV	0.5091	7.7521

SUMMARY STATISTICS FOR WEIGHT

BARTLETT TEST FOR HOMOGENEITY OF GROUP VARIANCES

CHI-SQUARE = 37.0695 DF= 11 PROBABILITY = 0.0001

ANALYSIS OF VARIANCE

SOURCE	SUM OF SQUARES	DF	MEAN SQUARE	F	PROBABILITY
BETWEEN GROUPS	6.5797	11	0.5982	3.5119	0.0002
WITHIN GROUPS	35.9381	211	0.1703		

SUMMARY STATISTICS FOR LENGTH

BARTLETT TEST FOR HOMOGENEITY OF GROUP VARIANCES

CHI-SQUARE = 91.0994 DF= 11 PROBABILITY = 0.0000

ANALYSIS OF VARIANCE

SOURCE	SUM OF SQUARES	DF	MEAN SQUARE	F	PROBABILITY
BETWEEN GROUPS	2667.4483	11	242.4953	9.9523	0.0000
WITHIN GROUPS	5141.1795	211	24.3658		

KOLMOGOROV-SMIRNOV ONE SAMPLE TEST USING STANDARD NORMAL DISTRIBUTION

VARIABLE	N-OF-CASES	MAXDIF	PROBABILITY (2-TAIL)
WEIGHT	223.0000	0.8686	0.0000
LENGTH	223.0000	1.0000	0.0000

LEVELS ENCOUNTERED DURING PROCESSING ARE:

TRT	1.000	2.000	3.000	4.000	5.000	6.000
REP	1.000	2.000				

DEP VAR: LENGTH N: 223 MULTIPLE R: 0.584 SQUARED MULTIPLE R: 0.342

ANALYSIS OF VARIANCE

SOURCE	SUM-OF-SQUARES	DF	MEAN-SQUARE	F-RATIO	P
TRT	2613.728	5	522.746	21.454	0.000
REP	1.709	1	1.709	0.070	0.791
TRT*REP	57.320	5	11.464	0.470	0.798
ERROR	5141.180	211	24.366		

Post-hoc contrast of treatment 1 with control.
TEST FOR EFFECT CALLED: TRT
TEST OF HYPOTHESIS

SOURCE	SS	DF	MS	F	P
HYPOTHESIS	16.390	1	16.390	0.673	0.413
ERROR	5141.180	211	24.366		

Post-hoc contrast of treatment 2 with control.
TEST FOR EFFECT CALLED: TRTTEST OF HYPOTHESIS

SOURCE	SS	DF	MS	F	P
HYPOTHESIS	135.223	1	135.223	5.550	0.019
ERROR	5141.180	211	24.366		

Post-hoc contrast of treatment 3 with control.
TEST FOR EFFECT CALLED: TRT
TEST OF HYPOTHESIS

SOURCE	SS	DF	MS	F	P
HYPOTHESIS	16.579	1	16.579	0.680	0.410
ERROR	5141.180	211	24.366		

Post-hoc contrast of treatment 4 with control.
TEST FOR EFFECT CALLED: TRT
TEST OF HYPOTHESIS

SOURCE	SS	DF	MS	F	P
HYPOTHESIS	121.446	1	121.446	4.984	0.027
ERROR	5141.180	211	24.366		

Post-hoc contrast of treatment 5 with control.
TEST FOR EFFECT CALLED: TRT
TEST OF HYPOTHESIS

SOURCE	SS	DF	MS	F	P
HYPOTHESIS	2062.126	1	2062.126	84.632	0.000
ERROR	5141.180	211	24.366		

LEVELS ENCOUNTERED DURING PROCESSING ARE:
TRT 1.000 2.000 3.000 4.000 5.000 6.000
REP 1.000 2.000

DEP VAR: WEIGHT N: 223 MULTIPLE R: 0.393 SQUARED MULTIPLE R: 0.155

ANALYSIS OF VARIANCE

SOURCE	SUM-OF-SQUARES	DF	MEAN-SQUARE	F-RATIO	P
TRT	6.353	5	1.271	7.460	0.000
REP	0.012	1	0.012	0.073	0.788
TRT*REP	0.240	5	0.048	0.282	0.923
ERROR	35.938	211	0.170		

Post-hoc contrast of treatment 1 with control.
TEST FOR EFFECT CALLED: TRT
TEST OF HYPOTHESIS

SOURCE	SS	DF	MS	F	P
HYPOTHESIS	0.276	1	0.276	1.618	0.205
ERROR	35.938	211	0.170		

Post-hoc contrast of treatment 2 with control.
TEST FOR EFFECT CALLED: TRT
TEST OF HYPOTHESIS

SOURCE	SS	DF	MS	F	P
HYPOTHESIS	0.132	1	0.132	0.772	0.381
ERROR	35.938	211	0.170		

Post-hoc contrast of treatment 3 with control.
TEST FOR EFFECT CALLED: TRT
TEST OF HYPOTHESIS

SOURCE	SS	DF	MS	F	P
HYPOTHESIS	0.095	1	0.095	0.559	0.455
ERROR	35.938	211	0.170		

Post-hoc contrast of treatment 4 with control.
TEST FOR EFFECT CALLED: TRT
TEST OF HYPOTHESIS

SOURCE	SS	DF	MS	F	P
HYPOTHESIS	0.338	1	0.338	1.983	0.161
ERROR	35.938	211	0.170		

Post-hoc contrast of treatment 5 with control.
TEST FOR EFFECT CALLED: TRT
TEST OF HYPOTHESIS

SOURCE	SS	DF	MS	F	P
HYPOTHESIS	5.287	1	5.287	31.042	0.000
ERROR	35.938	211	0.170		

Ecological Effects Branch One-Liner Data Entry Form

Chemical Fipronil and derivatives EUP (M&B 46030) Shaughnessy No. 129121 Pesticide Use _

AQUATIC VERTEBRATE TOX.	% AI	LC_{50} (95%CL) SLOPE	HRS/ TYPE	NOEC	STUDY/ REVIEW DATES	MRID/ CATEGORY	LAB	RC
1.								
2.								
3.								
4.								
5.								
6.								
7.								
CHRONIC TOX.	**% AI**	**MATC**	**DAYS**	**AFFECTED PARA.**	**STUDY/ REVIEW DATES**	**MRID/ CATEGORY**	**LAB**	**RC**
1. *Oncorhynchus mykiss*	96.7%	>6.6 <15 µg ai/l	90	length	1992/ 1994	429186-27 Core	SLI	RGM
2.								
3.								

COMMENTS: Results based on mean measured concentrations. SLI=Springborn Laboratories, Inc.

Fipronil Revised

Page _____ is not included in this copy.

Pages 23 through 26 are not included in this copy.

The material not included contains the following type of information:

_____ Identity of product inert ingredients.

_____ Identity of product impurities.

_____ Description of the product manufacturing process.

_____ Description of quality control procedures.

_____ Identity of the source of product ingredients.

_____ Sales or other commercial/financial information.

_____ A draft product label.

_____ The product confidential statement of formula.

_____ Information about a pending registration action.

✓ FIFRA registration data.

_____ The document is a duplicate of page(s) _____.

_____ The document is not responsive to the request.

The information not included is generally considered confidential by product registrants. If you have any questions, please contact the individual who prepared the response to your request.