DATA EVALUATION RECORD

CYCLOPROPENE, 1-METHYL-
(AFxRD-038)

STUDY TYPE: Nontarget Insect Testing (OPPTS 850.4350)

MRID 47088617

Prepared for
Biopesticides and Pollution Prevention Division
Office of Pesticide Programs
U.S. Environmental Protection Agency
One Potomac Yard
2777 South Crystal Drive
Arlington, VA 22202

Prepared by
Toxicology and Hazard Assessment Group
Environmental Sciences Division
Oak Ridge National Laboratory
Oak Ridge, TN 37830
Task Order No. 07-042

Primary Reviewer:
Eric B. Lewis, M.S. Signature:_____
Date: _____

Secondary Reviewers:
Anthony Q. Armstrong, M.S. Signature:_____
Date: _____

Robert H. Ross, M.S., Group Leader Signature:_____
Date: _____

Quality Assurance:
Susan S. Chang, M.S. Signature:_____
Date: _____

Disclaimer

This review may have been altered subsequent to the contractor=s signatures above.

Oak Ridge National Laboratory managed and operated by UT-Battelle, LLC., for the U.S. Department of Energy under Contract No. DE-AC05-00OR22725.

DATA EVALUATION RECORD

EPA Secondary Reviewer: Manying Xue, Chemist, 10/03/2007

STUDY TYPE:	Nontarget Insect Testing (OPPTS 850.4350)
MRID NO:	47088617
DP BARCODE:	DP339928
DECISION NO:	373522
SUBMISSION NO:	Not provided
TEST MATERIAL:	AFxRD-038 (a.i., 3.8% 1-methylcyclopropene)
STUDY NO:	Springborn Labs Study No. 1007.078.268
SPONSOR:	AgroFresh Inc., a Rohm and Haas Company, 100 Independence Mall, Philadelphia, PA 19106-2399
TESTING FACILITY:	Springborn Laboratories (Europe) AG, Seestrasse 21, Postfach, CH-9326 Horn, Switzerland
TITLE OF REPORT:	1-Methylcyclopropene: Laboratory Toxicity Test with the Predatory Mite, *Typhlodromus pyri* Scheuten (Acari: Phytoseiidae)
AUTHOR:	Nienstedt, K.M.
STUDY COMPLETED:	April 15, 2002
CONFIDENTIALITY CLAIMS:	None
GOOD LABORATORY PRACTICE:	A signed and dated GLP statement was provided. The study was conducted in compliance with the Swiss Ordinance relating to Good Laboratory Practice (2000), which is based on the OECD GLP. Stability, characterization, and verification of the test material identity and maintenance of records for the test material were the responsibility of the study sponsor.
STUDY SUMMARY:	A laboratory bioassay was conducted to determine the effects of a nominal atmospheric concentration of 10% 1-methylcyclopropene on the survival and reproduction of the predatory mite *Typhlodromus pyr*i. The test also included an α-cyclodextrin carrier control group and an untreated control group. The mites were exposed in closed containers for seven days, then removed and observed for reproduction for an additional seven days. There were no statistically significant differences in the survival or reproduction of the mites in the treated group compared to the control groups.

CLASSIFICATION: Acceptable

Test Material

3.4% 1-Methylcyclopropene alpha cyclodextrin complex, Lot No. BAS-5-80, TD No. 00-103, a white powder supplied by the study sponsor with an expiration date of January, 2009. A certificate of analysis was provided on p. 84 of MRID 47088617. After receipt at the testing facility, the test material was stored in the dark at room temperature.

The carrier control was Cavamax W6 Pharma, supplied by Rohm and Hass Company, Spring House, PA, Lot No. 60P300.

Test Methods

A laboratory study was conducted to determine the effects of the test material on the survival and reproduction of the predatory mite *Typhlodromus pyr*i. The test organisms were originally obtained from BASF, Limburgerhof, Germany, as eggs and maintained at the test facility under environmental conditions similar to the test conditions. For the test, an egg cohort was left undisturbed until protonymphs were present (4-5 days). At test start, the protonymphs were <24 hours old.

The test included a test material group, an α-cyclodextrin carrier control group, and an untreated control group. Each group consisted of four replicates of 20 mites each.

The test chambers were 15.4-cm diameter glass vessels with a total volume of four liters. The chambers were covered by glass lids containing a silicone septum in the center. Twenty test mites were placed onto a new glass cover slide (24 x 60 mm) containing a sticky barrier (Tangle-Trap Insect Trap Coating) along the edge, and food was placed on the slide (the food was not identified, but the test protocol specifies apple and walnut pollen). The slide was then placed onto a layer of black plastic and put in a glass Petri dish, which in turn was placed in the test chamber. No water was provided, since the humidity in the test vessels was high enough to prevent water stress. The test chambers were maintained in an environmental chamber at 25±2ºC with a photoperiod of 16 hours light:8 hours darkness.

Prior to placing the lid on the chamber, 2.7 mg of the test material or the carrier control material was weighed into a 3-cm x 3-cm glass vessel, which was hung by an inox wire inserted through the septum in the lid. The lid was then placed on the chamber and sealed by a metal cuff and silicone seal. After the chamber was sealed, a syringe containing 5 mL of Milli-Q water was inserted through the septum and discharged into the small glass vessel containing the test material powder, resulting in the production of 1-methylcyclopropene gas. The nominal concentration of the test material (2.7 mg/0.005 L) used was equivalent to 540 mg/L, or approximately 17.8 mg of active ingredient (calculated based on an active ingredient content of 3.3%). That concentration was selected based on calculations indicating that 17.8 mg of a.i./L would be equivalent to a 10 ppm (v/v) atmospheric concentration.

Additionally, quality control replicates were prepared to verify the dosing. On day 0, replicates were prepared by weighing 0.6 and 2.7 mg of the test material into weighing flasks, placing the

flasks in separate test vessels, and injecting them with 5 mL of Milli-Q water. On day 4, quality control replicates containing 0.65 and 2.74 mg of the test material were similarly prepared.

The initial treatments were repeated on test days 2 and 4 after the test chambers were opened and the mites were removed for observation. The number of living, dead, and missing mites, as well as the number stuck in the barrier, was recorded. After observation, survivors were transferred to a new cover slide with fresh food and returned to the test chambers. The chambers were then sealed and new treatment applications were performed.

On day 7, the test chambers were opened for observation, after which the mites were transferred to new test units and observed for reproduction during the following seven days. The new test units consisted of two side-by-side 24 x 60 mm glass cover slides with the middle seam joined by tape. A sticky barrier was placed around the outside edge. A layer of cotton soaked with deionized water was covered with black polyethylene foil and the slides were placed on top of the foil. A strip of tissue paper connected the top of the slides to the wet cotton layer. Fresh food was added to the slides on days 7, 9, and 11, when the number of surviving males and females, eggs, and larvae was recorded. Eggs and larvae were removed after each observation.

Mortality was considered as the sum of dead and missing mites, and mites stuck in the barrier. Mites that died during or directly after transfer to the chambers were not included in calculations of mortality. Mortality data were analyzed using a one-sided Fisher's exact test. Since the carrier control and the negative control were not significantly different, the test material treatment was compared to the carrier control. Mortality in the test material group was corrected for carrier control mortality using the Schneider-Orelli formula. Reproduction data were tested for normality using the Kolmogorov-Smirnov test and analyzed for significance using a t-test. The level of significance for all statistical comparisons was 5%.

On test days 0 and 4, samples of the atmosphere in all the treated vessels and one vessel in each control group were collected approximately four to six hours following addition of the water to determine the level of 1-methylcyclopropene in the test vessels. On test day 1 (approximately 23 to 30 hours after the addition of water on day 0), a sample was collected from all the treatment and control vessels. No samples were collected following the day 2 application. On day 7 samples were taken from all the test and control vessels, as well as the quality control vessels. Sampling was performed using a VICI VALCO Gas-tight A-2 syringe. Analysis was by gas chromatography with flame ionization detection. The method is detailed in pp. 55-65 of MRID 47088617. The instrument was a TRACE GC 2000 Series with a J&W Scientific GS-GASPRO column (0.32 mm id x 23 m). Isobutylene gas was used as the analytical standard.

Results Summary

During the test, the temperature ranged from 24.0 to 26.0ºC. In the closed test vessels during the exposure phase of the test the relative humidity was 100%. In the reproduction phase of the test, relative humidity ranged from 73% to 82%.

Mean recovery of 1-methylcyclopropene in the test material vessels ranged from 64.1 to 99.3% of the nominal concentration of 10 ppm during the test, with an overall mean of 90.3 ±17.5%. Mean recovery in the quality control vessels ranged from 57.4 to 98.5% of nominal, with an overall mean of 82.0 ±17.8%. No 1-methylcyclopropene was detected in the negative control or carrier control vessels.

Cumulative mortality of the mites is given in Table 1. There was no statistically significant difference in mortality between the negative control and carrier control groups, or between the test material and carrier control groups.

TABLE 1. Cumulative mortality of *T. pyri* exposed to 10 ppm 1-methylcyclopropene

Treatment	n[a]	Mean cumulative mortality (%)			Corrected mortality (%)[b]
		Day 2	Day 4	Day 7	
Negative control (0 ppm)	78	7.5 ± 5.0	16.7 ± 2.9	18.0 ± 5.5	-4.1
Carrier control (0 ppm)	74	2.5 ± 5.0	13.8 ± 4.8	21.3 ± 8.5	--
1-Methylcyclopropene (10 ppm)	80	3.8 ± 7.5	8.0 ± 6.3	15.4 ± 6.2	-7.5

Data from p. 26, MRID 47088617
[a]Variable numbers due to loss of mites during transfer to new test units
[b]Corrected for carrier control mortality, using Schneider-Orelli formula

Reproduction by the female mites is given in Table 2. There was no statistically significant difference in the number of eggs produced by females of the negative control and carrier control groups, or by the test material and carrier control groups.

TABLE 2. Reproduction per *T. pyri* female exposed to 10 ppm 1-methylcyclopropene

Treatment	Mean number of eggs laid per female				
	Day 7	Day 9	Day 11	Day 14	Total
Negative control (0 ppm)	0 ± 0	0.9 ± 0.5	2.5 ± 0.3	3.5 ± 0.6	7.0 ± 0.3
Carrier control (0 ppm)	0 ± 0	1.4 ± 0.7	3.0 ± 1.0	3.3 ± 0.3	7.8 ± 1.4
1-Methylcyclopropene (10 ppm)	0 ± 0	1.9 ± 0.6	3.2 ± 0.4	3.7 ± 0.6	8.8 ± 0.4

Data from p. 27, MRID 47088617

Study Author's Conclusions

The study author concluded that an atmospheric concentration of 10 ppm 1-methylcyclopropene had no statistically significant effect on survival or reproduction of *T. pyri* mites.

Reviewer's Conclusion

The reviewer agrees with the study author's conclusions. The reviewer notes that the concentration of active ingredient used in the test (3.4%) is slightly lower than that specified on the product label (3.8%).