RRB-89-84

ACUTE TOXICITY OF ISOMERS OF THE PYRETHROID INSECTICIDE DELTAMETHRIN AND ITS MAJOR DEGRADATION PRODUCTS TO *Daphnia magna*

by
K.E. Day, R. James Maguire

Rivers Research Branch
National Water Research Institute
Canada Centre for Inland Waters
Burlington, Ontario, L7R 4A6

November 1989
NWRI Contribution #89-161

ABSTRACT

The acute toxicities (4-h, 24-h and 48-h) of eight stereoisomers and four major breakdown products of deltamethrin to D. magna were determined. Parent deltamethrin (isomer 1) was the most toxic, with EC_{50} values in the range 0.05-1.75 µg/L, concentrations which have been observed in the surface microlayer and subsurface water in field studies. The other three isomers (2-, 3- and 4-deltamethrin) were two- to tenfold less toxic than 1-deltamethrin. Isomers 1', 2',3' and 4' plus the four breakdown product studied displayed no significant toxicity. Since 1-deltamethrin can be converted in natural water to 2', 3- and 4'- deltamethrin, and since 3-deltamethrin is toxic to D. magna, this isomerization is only a partial detoxification step as far as some aquatic organisms are concerned.

KEYWORDS - Deltamethrin Isomers Metabolic products
Daphnia magna

RESUME

Nous avons déterminé la toxicité aigue (4 heures, 24 heures et 48 pour D. magna de huit stéréoisomères et de quatre produits de décomposition de la deltaméthrine. La deltaméthrine d'origine (isomère 1) était l'espèce la plus toxique, les EC_{50} variant de 0,05 à 1,75 m g/L, soit des concentrations qui ont été observées dans la micro-couche superficielle et dans les eaux souterraines au cours d'études effectuées in situ. Les trois autres isomères (2-, 3- et 4-deltaméthrine) étaient de deux à dix fois moins toxiques que la 1-deltaméthrine. Les isomères 1', 2', 3' et 4', ainsi que les quatre produits de décomposition étudiés ne sont pas véritablement toxiques. Comme la 1-deltaméthrine peut être transformée, dans les eaux naturelles, en 2'-, 3'- et 4'-deltaméthrine et comme la 3-deltaméthrine est toxique pour

D. magna, cette isomérisation ne constitue, dans le cas de certains organismes aquatiques, qu'une détoxication partielle.

MOTS CLÉS - deltaméthrine, isomères, produits métaboliques,
Daphnia magna

MANAGEMENT PERSPECTIVE

The acute toxicities (4-, 24-h and 48-h) of eight stereoisomers and four major breakdown products of deltamethrin to D. magna were determined. Parent deltamethrin (isomer 1) was the most toxic, with EC_{50} values in the range 0.05 - 1.75 µg/L, concentrations which have been observed in the surface microlayer and subsurface water in field studies. The other three isomers (2-,3- and 4-deltamethrin) were two- to tenfold less toxic than 1-deltamethrin. Isomers 1', 2', 3' and 4' plus the four breakdown products studied displayed no significant toxicity. Since 1-deltamethrin can be converted in natural water to 2'- and 3- and 4'deltamethrin, and since 3-deltamathrin is toxic to D. magna, this isomerization is only a partial detoxification step as far as some aquatic organisms are concerned.

PERSPECTIVE-GESTION

Nous avons déterminé la toxicité aigue (4 heures, 24 heures et 48 heures) pour D. magna de huit stéréoisomères et de quatre produits de décomposition de la deltaméthrine. La deltaméthrine d'origine (isomère 1) était l'espèce la plus toxique, les EC_{50} variant de 0,05 à 1,75 m g/L, soit des concentrations qui ont été observées dans la micro-couche superficielle et dans les eaux souterraines au cours d'études effectuées in situ. Les trois autres isomères (2-, 3- et 4-deltaméthrine) étaient de deux à dix fois moins toxiques que la 1-deltaméthrine. Les isomères 1', 2', 3' et 4', ainsi que les quatre produits de décomposition étudiés ne sont pas véritablement toxiques. Comme la 1-deltaméthrine peut être transformée, dans les eaux naturelles, en 2'-, 3'- et 4'-deltaméthrine et comme la 3-deltaméthrine est toxique pour

D. magna, cette isomérisation ne constitue, dans le cas de certains organismes aquatiques, qu'une détoxication partielle.

INTRODUCTION

The synethic pyrethroid insecticide deltamethrin [(S)- alpha-cyano-3-phenoxybenzyl (1R,3R)-cis-2,2-dimethyl-3-(2,2-dibromovinyl) cyclopropanecarboxylate] is registered in Canada under the trade name Decis to control insects pests in large acreage crops such as potatoes, cereals, tobacco and cotton. Like many other synthetic pyrethroids, deltamethrin has been shown to be extremely toxic to fish [1] and aquatic invertebrates, especially custaceans [2], in laboratory studies. However, it has been suggested that the impact of deltamethrin impinging on the surfaces of aquatic ecosystems during application to nearby crops is lessened due to the rapid disappearance of the chemical from the water column [3-5]. The major routes of degradation or dissipation of deltamethrin from water have been reported to be (i) chemical and photochemical conversion to (2+2')-deltamethrin isomers, (ii) hydrolysis with subsequent oxidation of products and (iii) partitioning into suspended solids, plants, sediment, and air [4,5].

There are eight possible stereoisomers of deltamethrin, designated as 1, 1', 2, 2', 3, 3', 4 and 4' [6]. The parent compound (1-deltamethrin), with its cis-1R, 3 R configuration about the cyclopropane ring and the S configuration for the cyano group at the benzylic carbon atom, has the highest insecticidal activity [7]. It is the only enantiomer present in the registered product, even after storage for three years [8]. 3- Deltamethrin is the only other isomer known to have insecticidal [7] and mammalian [6] toxicity, although it

appears to require the synergist S,S,S-tributyl phosphorothrithioate, to be considered toxic to mice [6].

There is no information on the toxicity of isomers of deltamethrin to aquatic organisms. The conversion of the parent compound to the (2+2') isomers in natural waters under field conditions as a significant transformation pathway necessitates the determination of this toxicity. In addition, deltamethrin has been shown to rapidly degrade (hours to days) under field conditions to (at least) four major breakdown products, namely 3-phenoxybenzaldehyde (PBald), 3-phenoxybenzyl alcohol (PBalc), 3- phenoxybenzoic acid (PBacid) and cis-3-(2,2-dibromovinyl)-2,2-dimethylcyclopropane cabonxylic acid (DBCA) [4,5]. Stratton and Corke [9] found the PBald, PBalc and PBacid to be 3 - 50 times more effective than the parent insecticide permethrin at inhibiting photosynthesis and acetylene reduction in algae. The toxicity of these compounds to other aquatic organisms, particularly invertebrates, has not been reported.

The objective of this research was to determine the acute toxicity of the stereoisomers and breakdown products of deltamethrin to Daphnia magna.

MATERIAL AND METHODS

Analytical standards of all eight deltamethrin stereoisomers and DBCA were provided by Roussel Uclaf, Paris, France. The purity of the isomers was greater than 95% with the exception of the 2' isomer which was contaminated with about 15% of the 1' isomer. 3-Phenoxybenzaldehyde (PBald), 3-phenoxybenzyl alcohol (PBalc), and 3-phenoxybenzoic

acid (PBacid) were obtained from Aldrich Chemical Co., Milwaukee, WI. Analysis of all compounds was by chiral high performance liquid chromatography (HPLC) [8].

Stock concentrations of each isomer (5 mg/L) and degradation product (10 mg/L) were made by dilutions of the analytical standards in pesticide grade acetone (Caledon Laboratories, Georgetown, Ontario). Test concentration (0.025 - 25 or 50 or 382 µg/L) were prepared by serial dilution of the stock concentrations in filtered (0.45 um Whatman GFC) Lake Ontario water and were nominal. Controls were prepared by the addition of acetone to water at the highest concentration used in the bioassay experiments (i.e., 0.5%). Mortality in controls was never greater than 12% and was often 0%.

Daphnia magna were obtained from a culture maintained at the Canada Centre for Inland Waters, Burlington, Ontario. Gravid adult females were acclimated to experimental conditions (20-21°C; 16 hL:8D light regime) in 10L aquaria in filtered Lake Ontario water and fed ad libitum with the alga, Scendesmus spp., until neonates were produced. Groups of 5 young D. magna (<24h) were exposed to 200 mL of the test concentration in 250 mL glass beakers. Each concentration was replicated 3 times and each bioassay with the exception of isomer 4 was repeated twice. Efforts were made to disturb the animals as little as possible during handling and any that were damaged were discarded.

Percent mortality in terms of immobilization was determined at 4h, 24h, and 48h, and EC_{50} values were calculated by probit analysis using Parastat, a program developed and assembled by T. James at the University of Guelph, Guelph, Ontario, for the IBM personal computer.

Immobilization is defined as the inability of test animals to swim during a 10 sec. period of observation after the water in each beaker was gently swirled.

RESULTS AND DISCUSSION

Only isomers 1, 2, 3 and 4 are toxic to young D. magna (Table 1). The 1' 2', 3' and 4' isomers were not toxic at the highest concentrations tested. Of the four isomers which were toxic, the parent compound 1-deltamethrin was the most toxic. The acute toxicities of isomers 2, 3 and 4 were reduced by factors of approximately 2-10 compared to the parent isomer and for a 4 h exposure can be ranked in the decreasing order of toxicity 2 > 3 > 4. The immobilization levels reported here indicate that parent 1-deltamethrin is the most toxic to D. magna of four synthetic pyrethroids currently registered for agricultural use (e.g., 24-48 h EC_{50} values are 1.0 - 5.0 µg/L for cypermethrin, 0.2 - 2.1 µg/L for permethrin and 0.83 - 2.1 µg/l for fenvalerate compared to 0.05-0.29 µg/L for deltamethrin) [10]. Since 1-deltamethrin has been shown to isomerize over a period of hours to days [5], more confidence is place on EC_{50} values obtained at 4 h than those obtained at longer time intervals.

The toxicities of the eight stereoisomers of deltamethrin to biota have been shown to vary considerably, with only isomer 1 (1R, 3R, alpha S) and to a lesser extent isomer 3 (1R, 3S, alpha S), being highly active in insects and mammals [6,7]. The present study is the first to report the toxicity of isomer 2 (1R,3R, alpha R), isomer 3

and isomer 4 (1R, 3S, alpha R) to the aquatic invertebrate, D. magna. The importance of stereoisomerism as a factor influencing the activity of pyrethroids is well established [12]. The results from this study indicate that the target site for deltamethrin in crustaceans such as D. magna appears to have less rigid sterospecific requirements that for other arthropods.

There have been very few determinations of isomers of pyrethroid insecticides in natural waters under field conditions. Using an achiral method of analysis, Maguire et al. [5] reported on the concentrations of the four pairs of deltamethrin enantiomers (1+1', 2+2', 3+3', and 4+4') in a pond directly oversprayed with deltamethrin in Prince Edward Island, Canada. Concentrations of (1+1')-deltamethrin in the surface microlayer sampled immediately following aerial application of the chemical were about 50 µg/L, substantially higher than the 4 h EC_{50} value of 1.75 µg/L, determined in this work. The half-life of these isomers in water was about 5 min, but 0.042 µg/L remained 55 h after spray, a concentration which is close to the 48 h EC_{50} value for 1-deltamethrin. Initial concentrations of (1+1')-deltamethrin in subsurface waters were much less than those reported in the surface microlayer, approximtely 0.320 µg/L. The half-life of dissipation of (1+1')-deltamethrin in subsurface water was about 1 h. It has since been found by chiral analysis that the (1+1') isomer in water is solely the 1 isomer [8]. These results indicate that concentrations of 1-deltamethrin applied directly to natural waters are high enough and remain in the water column long enough to be acutely toxic to cladocerans even though they disappear relatively

quickly following application. Tooby et al. [3] found that concentrations in subsurface water estimated to range from 0.75 to 1.0 µg/L total deltamethrin severly depleted insects and crustaceans in ponds sprayed at normal field application levels. Even when a 10 m or 100 m buffer zone was imposed on ground or aerial spray regimes to protect the aquatic invironment, Ernst et al. [13] found that surface water concentrations of deltamethrin in the setback ponds were still in the range of 0.01 - 0.05 µg/L, concentrations that could be toxic to D. magna if maintained for 48 h or more.

Relatively large concentrations of the (2+2') isomers were also found in the pond study by Maguire et al. [5] in both the subsurface water (0.014 µg/L) and in the surface microlayer (6.1 µg/L) shortly after spray. Over 1-3 d, this pair of isomers became a major contributor to the total deltamethrin concentration. For example, at 25 h after the spray its concentration was 69% that of the (1+1') isomer. These concentrations could be considered detrimental to the health of cladocerans if they represented only the 2 isomer. However, a subsequent chiral analysis has determined that 1-detramethrin in natural water undergoes a dark chemical conversion to 2'-deltamethrin, an isomer found to be non-toxic to D. magna in the laboratory (Table 1). No 2-deltramethrin is produced. These results suggest that concentrations of (2+2')-deltamethrin determined in the earlier pond study [15] probably represented only the 2' isomer which is not toxic.

1-Deltamethrin in natural water can also be converted to 3- and 4'-deltamethrin, in addition to 2'-deltamethrin, through sunlight photolysis [8]. Although these products were not found in the

earlier pond study [5], the enantiometic pairs (3+3')- and (4+4')-deltamathrin in addition to (2+2')-deltamethrin, have been observed on pasture forage and litter [14] and alfalfa]15]. The data in Table 1 indicate that while 4'-deltamethrin is not toxic to D. magna, conversion of parent 1-deltamethrin to 3-deltamethrin is only a partial detoxification step.

All four major degradation products of deltamethrin (DBCA, PBald, PBalc, PBacid) have been found in the surface microlayer and subsurface water of ponds in concentrations as high as 0.08 µg/L 24 h after the application of 1-deltamethrin [4,5]. These concentrations and those considerably higher (up to 50 µg/L) were found to be non-toxic over 48 h to D. magna in the present study. Metabolites of the synthetic pyrethroids have generally been found to be less toxic in animals [6]. However, Stratton and Corke [9] found that two degradation products of permethrin, PBalc and PBald, were 3 - 50 times more effective than the parent compound in inhibiting photosynthesis and acetylene reduction in algae and cyanobacteria at concentrations 100 - 1000 times higher than those used in the present study. The cyanbacteria were more sensitive than the green algae and the authors attributed this to the basic cellular organizational differences between these organisms (procaryotic vs. eucaryotic cells). Higher concentrations of metabolites were not tested in the present study for their toxicities to D. magna, since such concentrations are unlikely to be present under realistic field conditions.

ACKNOWLEDGEMENTS

The authors thank Mr. Ian M. Scott for his capable assistance in the laboratory. Samples of the stereoisomers of deltamethrin were provided by Dr. J. Tessier of Roussel Uclaf, Paris, France and are gratefully acknowledged.

REFERENCES

1. Mulla, M.S., Navvab-Gojrati, H.A., Darwazeh, H.A., 1978. Toxicity of Mosquito Larvicidal Pyrethroids to Four Species of Freshwater Fishes. Environ. Entomol. 7: 428-430.

2. Zitko, V., McLeese, D.W., Metcalfe, C.D., and Carson, W.G., 1979. Toxicity of Permethrin, Decamethrin and Related Pyrethroids to Salmon and Lobster. Bull. Environ. Contam. Toxicol. 21: 338-343.

3. Tooby, T.E., Thompson, A.N., Rycroft, R.O., Black, I.A., and Hewson, R.T., 1981. A Pond Study to Investigate the Effects on Fish and Aquatic Invertebrates of Deltamethrin Applied Directly to Water. U.K. Ministry of Agriculture, Fisheries and Food, Burnham-on-Crouch, Essex, England, 25 pp.

4. Muir, D.C.G., Rawn, G.P., and Grift, N.P. 1985. Fate of the Pyrethroid Insecticide Deltamethrin in Small Ponds: A mass balance study. J. Agric. Food Chem. 33: 603-609.

5. Maguire, R.J., Carey, J.H., Hart, R.H., Tkacz, R.J., and Lee, H.-B., 1989. Persistance of Fate of Deltamethrin Sprayed on a Pond. J. Agric. Food Chem. 37: 1153-1159.

6. Ruzo, L.O., Holmstead, R.L., Casida, J.E., 1977. Pyrethroid Photochemistry: decamethrin. J. Agric. Food Chem. 1385-1394.

7. Tessier, J., 1982 Structure, Synthesis and Physical-chemical Properties of Deltamethrin. In Deltamethrin Monograph. J. Lhoste, Ed., roussel Uclaf, Paris, France, pp. 37-66.

8. Maguire, R.J., Chemical, and Photochemical Isomerization of Deltamethrin. Submitted for publication.

9. Stratton, G.W., and Corke, C.T., 1982. Toxicity of the Insecticide Permethrin and Some Degradation Products Towards Algae and Cyanobacteria. Environ. Poll. (Ser. A) 29: 71-80.

10. Day, K.E., 1989. Acute, Chronic anc Sublethal Effects of Synthetic Pyrethroids on Freshwater Zooplankton. Environ. Toxicol. Chem. 8: 411-416.

11. Elliott, M., Farnham, A.W., Janes, N.F., Needham, P.H., Pullman, D.A., 1974. Synthetic Insecticide With a New Order of Activity. Nature§ 248: 710-711.

12. Elliott, M., and Janes, N.R., 1977. Preferred Conformations of Pyrethroids. In Synthetic Pyrethroids, M. Elliott, American Chemical Society Symposium Series 42, American Chemical Society, Washington, D.C., U.S.A., pp 29-36.

13. Ernst, W.R., Maguire, R.J., Carey, J.H., Julien, G.R., Hart, J.H. Tkacz, R.J., Lee, H.-B., 1989. The Fate, Persistence and Biological Impacts of Deltamethrin Applied Directly to an Agricultural Pond System and the Protection from Deposit Afforded by a 100 Metre Set-back. Department of Environment, Environmental Protection - Atlantic Region Surveillance Report EPS-5-AR-89-1, Dartmouth, Nova Scotia, Canada, 57+ pp.

14. Hill, B.D., Johnson, D.L., 1987. Persistence of Deltamethrin and its Isomers on Pasture Forage and Litter. J. Agric. Food. Chem. 35: 373-378.

15. Hill, B.D., Inaba, D.J., and Charnetski, W.A., 1989. Dissipation of Deltamethrin applied to Forage Alfalfa. J. Agric. Food Chem. 37: 1150-1153.

TABLE 1 Toxicity of isomers and breakdown products of Deltamethrin to Daphnia magna.

Isomer	Time	EC-50[a] (µg/L)	Slope of probit Line
1	4	1.75 (0.94-3.27)[b]	0.73
	24	0.27 (0.09-0.84)	0.40
		0.29 (0.18-0.45)	1.14
	48	0.07 (0.06-0.09)	3.41
		0.05 (0.03-0.09)	0.87
1'	4	> 25	
	24	> 25	
	48	> 25	
2	4	5.04 (2.84-8.97)	1.11
	24	0.50 (0.31-0.78)	1.27
		0.47 (0.22-0.39)	0.62
	48	0.27 (0.19-0.39)	1.60
		0.13 (0.06-0.28)	0.59
2'	4	> 25	
	24	> 25	
	48	> 25	
3	4	7.45 (5.32-10.43)	1.98
	24	2.22 (0.90-5.51)	0.70
		1.85 (0.96-3.57)	0.73
	48	0.37 (0.23-0.60)	1.17
		0.60 (0.34-1.04)	0.84
3'	4	> 382	
	24	> 382	
	48	> 382	
4	4	>25	
	24	2.54 (1.16-5.57)	0.62
	48	0.22 (0.14-0.36)	1.05

continued on next page

TABLE 1 (continued) Toxicity of isomers and breakdown products of Deltamethrin to Daphnia magna.

Isomer	Time	EC-50[a] (µg/L)	Slope of probit Line
4'	4	> 25	
	24	> 25	
	48	> 25	
PB alc	24	> 50	
	48	> 50	
PB ald	24	> 50	
	48	> 50	
PB acid	24	> 50	
	48	> 50	
DBCA	24	> 50	
	48	> 50	

[a] Maximum likelihood estimate

[b] Numbers in parentheses are minimum and maximum EC-50 values predicted by probit analysis