Letsinger, R. L., Steller, K. E., *Tetrahedron Lett.*, 1401 (1969).
Lutz, R. E., Allison, R. K., Ashburn, G., Bailey, P. S., Clark, M. T., Codington, J. F., Deinet, A. J., Freek, J. A., Jordan, R. H., Leake, N. H., Martin, T. A., Nicodemus, K. C., Rowlett, R. J., Jr., Shearer, N. H., Jr., Smith, J. D., Wilson, J. W., III, *J. Org. Chem.* **12**, 617 (1947).
Nakagawa, M., Crosby, D. G., *J. Agr. Food Chem.* **22**, 849 (1974).
Ogata, Y., Takagi, K., Ishino, I., *Tetrahedron* **26**, 2703 (1970).
Plimmer, J. R., Klingebiel, U. I., *Science* **174**, 407 (1971).
Ross, W. C. J., "Biological Alkylating Agents," Butterworth, London, 1962.
Steller, K. E., Letsinger, R. L., *J. Org. Chem.* **35**, 308 (1970).
Traylor, T. G., Bartlett, P. D., *Tetrahedron Lett.*, 30 (1960).
Weed Science Society, "Herbicide Handbook of the Weed Science Society of America," 2nd ed, Humphrey Press, Geneva, N. Y., 1970, p 174.

Received for review November 6, 1972. Resubmitted July 17, 1974. Accepted July 27, 1974. Presented at the Division of Pesticide Chemistry, 162nd National Meeting of the American Chemical Society, Washington, D. C., Sept, 1971. Supported in part by Research Grant ES-00054 (U. S. Public Health Service) and Regional Research Project W-45 (U. S. Department of Agriculture).

Photodecomposition of Mirex on Silica Gel Chromatoplates Exposed to Natural and Artificial Light

G. Wayne Ivie,* H. Wyman Dorough, and Earl G. Alley

Exposure of mirex to sunlight or ultraviolet light as deposits on silica gel thin-layer chromatoplates resulted in its slow degradation to several products. Photoproducts characterized included Kepone hydrate, a monohydromirex derivative previously identified (1,2,3,4,5,5,6,7,9,10,10-undecachloropentacyclo[5.3.0.$0^{2,6}$.$0^{3,9}$.$0^{4,8}$]decane), and 1,2,3,4,6,7,9,10,10-nonachloropentacyclo[5.3.0.$0^{2,6}$.$0^{3,9}$.$0^{4,8}$]decan-5-one, isolated as the hydrate. Because certain of the mirex photoproducts are considerably more polar than either mirex or its monohydro derivative, any environmental degradation of mirex *via* these pathways would possibly result in enhanced biodegradability and a reduced tendency toward storage and accumulation in adipose tissues.

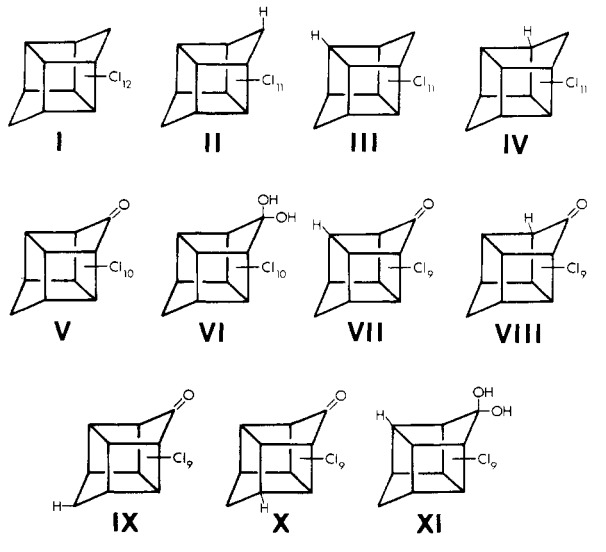
The insecticide mirex (dodecachloropentacyclo[5.3.0.$0^{2,6}$.$0^{3,9}$.$0^{4,8}$]decane (I)) is highly effective in controlling the imported fire ant (*Solenopsis spp.*), which infests large areas of the southeastern United States and extends westward into Texas. The compound is generally applied by aircraft as a bait formulation at rates of only a few grams of toxicant per acre to provide effective fire ant suppression.

Mirex may be an environmental hazard because it is highly toxic to certain aquatic organisms, particularly crustaceans (Ludke *et al.*, 1971), and is high in chronic toxicity to some fishes (Van Valin *et al.*, 1968). Mirex is low in acute toxicity to mammals (Gaines and Kimbrough, 1970; Martin, 1972), but it is an inducer of hepatic mixed function oxidases (Baker *et al.*, 1972; Mehendale *et al.*, 1973). Dietary mirex at levels as low as 5 ppm causes reduced litter size in mice (Ware and Good, 1967) and at 25 ppm causes cataracts in rats (Gaines and Kimbrough, 1970).

Studies to date have indicated that mirex is highly resistant to metabolic attack by higher organisms, including laboratory rats (Gibson *et al.*, 1972; Mehendale *et al.*, 1972), Japanese quail (Ivie *et al.*, 1974a), cattle (Dorough and Ivie, 1974), and plants (Mehendale *et al.*, 1972). More recent studies have indicated that mirex is metabolized very slowly by sewage sludge organisms under anaerobic conditions, and the single metabolite generated has been identified as the monohydro product II (1,2,3,4,5,5,6,7,8,9,10-undecachloropentacyclo[5.3.0.$0^{2,6}$.$0^{3,9}$.$0^{4,8}$]decane) (Andrade and Wheeler, 1973).

The metabolic stability of mirex and its highly lipophilic nature suggest that this chlorocarbon may show considerable potential for biological magnification through accumulation of residues in certain tissues of a variety of food chain organisms. Indeed, studies involving long-term administration of mirex to certain mammals, birds, and fish indicate that mirex is retained and accumulates at high levels in fatty tissue (Ivie *et al.*, 1974b).

Certain aspects of the photochemistry of mirex have been reported. Monohydro- and dihydromirex photoproducts were isolated after ultraviolet (uv) lamp irradiations of mirex in hydrocarbon solvents (Alley *et al.*, 1973), and the same monohydro derivative was observed after exposures of mirex deposits on silica gel surfaces (Gibson *et al.*, 1972). Definitive structure assignment of the monohydro product as 1,2,3,4,5,5,6,7,9,10,10-undecachloropentacyclo[5.3.0.$0^{2,6}$.$0^{3,9}$.$0^{4,8}$]decane (III) has recently been made (Alley and Layton, 1973). The third possible monohydro product of mirex, IV, has not been observed.



Veterinary Toxicology and Entomology Research Laboratory, U.S. Department of Agriculture, Agricultural Research Service, College Station, Texas 77840 (G.W.I.), Department of Entomology, University of Kentucky, Lexington, Kentucky 40506 (H.W.D.), Mississippi State Chemical Laboratory, Mississippi State, Mississippi 39762 (E.G.A.).

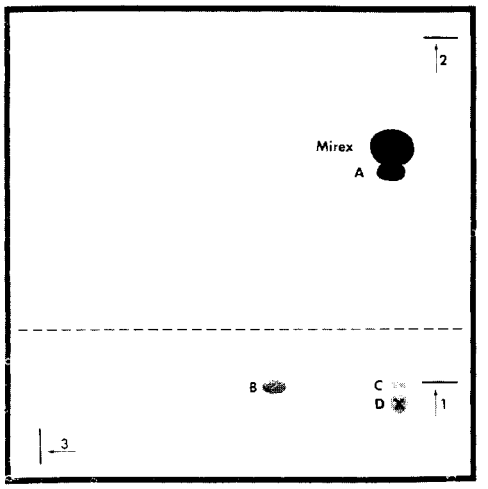


Figure 1. Drawing of a thin-layer radioautogram showing resolution of mirex-^{14}C photoproducts. Plates were developed about 1 cm above origin in methanol (solvent 1) to minimize binding of radiocarbon at the origin, then in heptane (solvent 2) to resolve mirex from photoproduct A. Plates were subsequently cut at dotted line and the lower portion developed in 2:1 ether–hexane (solvent 3) to resolve photoproduct B.

The studies reported here were undertaken to determine if exposure of mirex to sunlight might result in more readily biodegradable compounds, and to determine the chemical nature of any photoproducts generated. Because the photodecomposition of mirex occurs very slowly in sunlight, exposures were made as deposits on thin silica gel plates to give a stable support providing maximum area for surface exposure. Mirex deposits were also exposed to artificial light to permit isolation and characterization of products generated in sunlight.

MATERIALS AND METHODS

Chemicals. Uniformly labeled mirex-^{14}C (6.34 mCi/mmol) was obtained from Mallinckrodt Chemical Works, St. Louis, Mo. Analytical grade mirex and technical Kepone hydrate were obtained from Allied Chemical Corp., Baltimore, Md. The radiolabeled and unlabeled mirex samples gave only a single spot on thin-layer chromatography (tlc) in several solvent systems in which radioautography was used to detect the radioactive material and diphenylamine–ultraviolet light for visualization of the unlabeled compound. The mirex samples also gave single symmetrical peaks on gas–liquid chromatography (glc).

Analytical Procedures. Infrared (ir) spectra were recorded as 1% potassium bromide pellets on a Beckman IR-18A spectrophotometer. Mass spectra were determined with a Varian-MAT CH-7 90° sector magnetic scan spectrometer, with an ionizing voltage of 70 eV. The ion source temperature was maintained at 250°. Samples were analyzed either by direct insertion probe or by glc–mass spectral straight-line inlet coupling with a Varian 2700 chromatograph equipped with a 6 ft × ⅛ in. stainless-steel column containing 3% SE-30 on 100–120 mesh varaport 30. Operating parameters were as follows: injector, 215°; column, 200°; detector, 220°; helium carrier, 50 ml/min.

Sunlight Photodecomposition of Mirex-^{14}C. Methanol solutions of mirex-^{14}C were applied in one corner of 20 × 20 cm Eastman Chromagram Sheets (100 μ gel thickness, Type 6061, without fluorescent indicator) to give 6-μg deposits in circular spots 0.5–0.7 cm in diameter. The plates were secured to a large section of plywood and placed in direct summer sunlight at an angle toward the sun to obtain maximum exposure. Samples were left outside except during rain, and were analyzed periodically.

The plates described above were preferred to conventional glass-backed pour on or precoated tlc plates for several reasons. The flexible polyethylene terephthalate backing of the plates provides much better binding of the gel to the support; this is highly desirable for extended outdoor exposures. These plates stand up well to wetting by dew or accidental exposure to even heavy rainfall, whereas glass backed plates invariably peel under such conditions. The thin (100 μ) gel coating of these plates is preferred because sample exposure is more complete than with thicker coatings.

The photodegradation products were resolved by direct solvent development of the exposed chromagram sheets. The plates were initially developed in methanol until the solvent front had risen about 1–1.5 cm above the mirex spot. Preliminary experiments indicated that the radiocarbon tended to become tightly bound at the origin, especially on extended exposures, and the methanol predevelopment was effective in freeing this material for normal solvent development. After partial development in methanol, plates were dried, then developed in *n*-heptane to resolve mirex from its known monohydro product III (R_f values: mirex, 0.68; III, 0.61). Developed plates were cut with scissors in a horizontal line about 4 cm above the original mirex spot (Figure 1), and the lower part was developed in the second dimension in 2:1 ether–hexane for further photoproduct resolution. After development, the two sections were rejoined by taping the back surface, and the plates were exposed to X-ray film to visualize radioactive gel regions. These areas were then scraped and quantitated by liquid scintillation counting.

Isolation of Mirex Photoproducts. Mirex solutions (10 mg/ml of benzene) were streaked over 20 × 20 cm chromagram sheets to a concentration of about 10 mg/plate. The plates were exposed to Westinghouse fluorescent sunlamps at a distance of 10 cm for 7 days. These lamps emit uv radiation above 275 nm, with maximum energy output at 310 nm. The gel was scraped from the plates, ground with a mortar and pestle, and extracted with hexane to remove a mixture of the monohydro product III and unreacted mirex. III was isolated and purified as previously described (Gibson *et al.*, 1972). The gel was then extracted with ether to obtain the major polar photoproduct along with small amounts of residual mirex and III. The polar material was subsequently isolated by preparative silica gel tlc developed in 2:1 ether–hexane.

RESULTS AND DISCUSSION

Degradation of Mirex-^{14}C in Sunlight. Detectable levels of mirex photoproducts are observed within 3 days of sunlight exposure of mirex-^{14}C (Table I). Products A and B (Figure 1) predominate, whereas products C and D are generated in lower amounts. Mirex degradation under these parameters proceeds very slowly, and even after 28 days of exposure, almost 90% of the persisting residue consisted of unchanged mirex. Not reflected in Table I are radiocarbon losses from the plates through volatility or weathering of the gel itself. Such losses were negligible with the 3-day samples, but amounted to approximately 20% of the applied radiocarbon in samples analyzed after 4 weeks of exposure. Degradation of mirex was negligible when treated plates were held in the laboratory in darkness for 28 days (Table I).

Chemical Nature of Mirex Photoproducts A and B. The major photoproduct A produced after exposure of mirex to sunlight was identified as the monohydro derivative III. The compound was identical in glc and tlc comparisons with III isolated in milligram amounts after mirex exposure to uv light, and the spectral data obtained from III isolated after mirex exposures to uv light in these studies agreed with data in published reports (Alley *et al.*, 1973; Gibson *et al.*, 1972).

Mirex photoproduct B is more polar than either mirex or III. It does not migrate on tlc developed in heptane

Table I. Degradation of Mirex-^{14}C Exposed to Sunlight as Deposits on Silica Gel Thin-Layer Chromatograms

Exposure, days	Recovered radiocarbon as indicated product, %				
	Mirex	A	B[a]	C	D
0	99.8 ± 0.0	0	0	0.1 ± 0.0	0.1 ± 0.0
3	98.1 ± 0.2	1.0 ± 0.1	0.4 ± 0.0	0.2 ± 0.0	0.3 ± 0.0
7	96.5 ± 0.3	2.1 ± 0.2	0.6 ± 0.1	0.4 ± 0.0	0.4 ± 0.0
14	94.0 ± 0.6	3.7 ± 0.3	1.2 ± 0.1	0.6 ± 0.0	0.5 ± 0.0
28	88.1 ± 0.6	6.8 ± 0.1	3.6 ± 0.3	1.0 ± 0.1	0.5 ± 0.0
28 (dark)	98.9 ± 0.3	0.6 ± 0.4	0.2 ± 0.0	0.1 ± 0.0	0.1 ± 0.0

[a] This product consisted of a mixture of compounds VI and XI which were not resolved by tlc.

(Figure 1), but does move in 2:1 ether-hexane (R_f 0.35). Ir of the compound isolated after preparative scale exposures of mirex to uv light showed a strong broad band at 3500 cm^{-1}, characteristic of hydroxyl absorption, but the ir did not indicate the presence of other functional groups. Glc-mass spectral studies with photoproduct B indicated it to be only about 90% pure. The product consisted of two components; retention time was 3.6 min for the minor component and 5.1 for the major. The major component gave a small parent ion at *m/e* 486 ($C_{10}Cl_{10}O^+$), and the base peak was at *m/e* 270 ($C_5Cl_6^+$). The latter ion is characteristic of the pentacyclodecane nucleus and represents a major fragmentation mode in the mirex series (Dilling and Dilling, 1967; Dilling *et al.*, 1967). Other prominent fragments were at *m/e* 451 ($C_{10}Cl_9O^+$), 353 ($C_9Cl_7^+$), 283 ($C_9Cl_5^+$), 235 ($C_5Cl_5^+$), and 216 ($C_5Cl_4O)^+$. The mass spectral fragmentation patterns of this compound were consistent with its tentative assignment as Kepone (V), which is generated by photolytic replacement of the two chlorine atoms on one of the dichloromethylene carbons of mirex with a carbonyl group. The compound is then rapidly hydrated to the *gem*-diol VI. Chromatographic and spectral studies with authentic Kepone hydrate (VI) confirmed the assignment of photoproduct B as Kepone hydrate. The photoproduct behaved the same on tlc and glc as Kepone hydrate, and the ir and mass spectral data of the two compounds were essentially identical.

Glc-mass spectral studies of the minor component of photoproduct B discussed above revealed a small parent ion at *m/e* 452 ($C_{10}Cl_9HO^+$), a base peak at *m/e* 270 ($C_5Cl_6^+$), and other prominent ions at *m/e* 249 ($C_9Cl_4H^+$), 236 ($C_5Cl_5H^+$), 235 ($C_5Cl_5^+$), 216 ($C_5Cl_4O^+$), 201 ($C_5Cl_4H^+$), and 182 ($C_5Cl_3HO^+$). These fragmentation patterns of the product permitted its unambiguous assignment as the monohydrokepone analog VII, isolated as the hydrate XI. Of the four possible monohydrokepone isomers (VII-X) only VII can yield both the $C_5Cl_6^+$ and $C_5Cl_5H^+$ ions observed. Compound VII has also been identified as a Kepone photoproduct in earlier studies (Alley and Layton, 1973). Exposure of the purified monohydromirex photoproduct III on silica gel to artificial light resulted in the conversion of III to VII based on glc-mass spectral, and thus the very low levels of VII observed in these studies probably involved both Kepone and the monohydromirex photoproduct as precursors. The chemical nature of the very minor mirex photoproducts C and D was not investigated.

The present studies do not definitively establish that mirex will undergo significant photodegradation in the environment under natural exposure conditions. Indeed, the very slow decomposition observed under parameters designed for maximum exposure suggests that photolytic degradation of mirex in the environment will occur at an exceedingly slow rate. These studies do indicate, however, that photochemical energy in sunlight is capable of degrading mirex, perhaps the most environmentally persistent organic pesticide known, to products of a more polar nature that may well be more susceptible than mirex to biodegradation processes and less likely to accumulate in tissues.

ACKNOWLEDGMENT

For technical assistance, the authors thank Darcy Rushing at the Veterinary Toxicology and Entomology Research Laboratory and Jane Johnson at the University of Kentucky.

LITERATURE CITED

Alley, E. G., Dollar, D. A., Layton, B. R., Minyard, J. P., Jr., *J. Agr. Food Chem.* **21**, 138 (1973).
Alley, E. G., Layton, B. R., paper presented at the 165th National Meeting of the American Chemical Society, Dallas, Tex., April 9–13, 1973.
Andrade, P. S. L., Wheeler, W. B., paper presented at the 166th National Meeting of the American Chemical Society, Chicago, Ill., Aug 26–31, 1973.
Baker, R. C., Coons, L. B., Mailman, R. B., Hodgson, E., *Environ. Res.* **5**, 418 (1972).
Dilling, W. L., Braendling, H. P., McBee, E. T., *Tetrahedron* **23**, 1211 (1967).
Dilling, W. L., Dilling, M. L., *Tetrahedron* **23**, 1225 (1967).
Dorough, H. W., Ivie, G. W., *J. Environ. Qual.* **3**, 65 (1974).
Gaines, T. B., Kimbrough, R. D., *Arch. Environ. Health* **21**, 7 (1970).
Gibson, J. R., Ivie, G. W., Dorough, H. W., *J. Agr. Food Chem.* **20**, 1246 (1972).
Ivie, G. W., Dorough, H. W., Bryant, H. E., *Bull. Environ. Contam. Toxicol.* **11**, 129 (1974a).
Ivie, G. W., Gibson, J. R., Bryant, H. E., Begin, J. J., Barnett, J. R., Dorough, H. W., *J. Agr. Food Chem.* **22**, 646 (1974b).
Ludke, J. L., Finley, M. T., Lusk, C. I., *Bull. Environ. Contam. Toxicol.* **6**, 89 (1971).
Martin, H., Ed., "Pesticide Manual," British Crop Protection Council, 1972.
Mehendale, H. M., Chen, P. R., Fishbein, L., Matthews, H. B., *Arch. Environ. Contam. Toxicol.* **1**, 245 (1973).
Mehendale, H. M., Fishbein, L., Fields, M., Matthews, H. B., *Bull. Environ. Contam. Toxicol.* **8**, 200 (1972).
Van Valin, C. C., Andrews, A. K., Eller, L. L., *Amer. Fish Soc. Trans.* **97**, 185 (1968).
Ware, G. W., Good, E. W., *Toxicol. Appl. Pharmacol.* **10**, 54 (1967).

Received for review June 13, 1974. Accepted August 22, 1974. This research was initiated at the University of Kentucky and was supported in part by U.S. Department of Agriculture Cooperative Agreements No. 12-14-100-10,947 (33), at the University of Kentucky, and 12-14-100-10, 939 (33), at the Mississippi State Chemical Laboratory. Mention of a trade name, proprietary product, or specific equipment does not constitute a guarantee or warranty by the U.S. Department of Agriculture and does not imply its approval to the exclusion of other products that may be suitable.