Mirex Incorporation in the Environment: Phytotoxicity on Germination, Emergence, and Early Growth of Crop Seedlings¹

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In recent years, phytotoxic effects of pesticides and herbicides on seed germination, survival, and growth of seedlings have been reported by several investigators (e.g., COX and LILLY 1952; AMEEN et al. 1960; BOWLING 1964; OLIVER and FRANS 1968; DALVI et al. 1972; GAWAAD et al. 1972b). Laboratory and greenhouse studies by CHANG and FOY (1971) revealed that picloram (4-amino-3, 5, 6-trichloropicolinic acid) inhibited germination of soybean and safflower whereas radish and barley were not affected. COX and LILLY (1952) reported that germination and early growth of several field crops were affected by aldrin and dieldrin. CULLINAN (1949) stated that small quantities of DDT, BHC, chlorodane, and toxaphene, when applied to soil, depressed the growth of some crop seedlings. Similarly, GAWAAD et al. (1972a) observed that lindane was highly toxic to clover seedlings and suppressed growth, whereas the effects of DDT and heptachlor were very slight.

Mirex (dodecachloropentacyclo-1, 3, 4-metheno-2H-cyclobyta (cd) pentalene) was put into general field use in 1962 to control the imported fire ant (Solenopsis saevissima Forel, Formicidae). Mirex residue and toxicity to non-target organisms have recently received increasing attention (NAQVI and DE LA CRUZ 1973; DE LA CRUZ and NAQVI 1973). Little work has been done on the effect of this persistent insecticide on plants, specifically on seed germination, seedling emergence, survival and growth. It was observed that no mirex residue could be detected in several crops and grass grown in soil having residues of 0.01 ppm level (MARKIN et al. 1972) although bahiagrass (Paspalum notatum Flugge) contained 0.0003 to 0.017 ppm residues when grown in soil containing 0.001 to 0.002 ppm mirex. Using higher concentrations of mirex (0.3 to 3.5 ppm) we observed that seedlings of different crops take up and accumulate varying amounts of residues in different parts of the plant.

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The objective of this study is to determine the effects of mirex on germination, emergence, and growth of seedlings of several commonly grown field and pasture crops. In order to achieve the objective of this investigation we used mirex concentrations higher than what is naturally found in the environment.

MATERIALS AND METHODS

The six crops tested were crimson clover (Trifolium incarnatum L. Var: elatius Gibelli and Bali.), johnson grass (Sorghum halpense (L.) Pers.), annual rye grass (Lolium multiflorum Lam.), tall fescue (Festuca arundunacea. Var: 'Alta'), alsike clover (Trifolium hybridum L.) and alfalfa (Medicago sativa L. Var: 'Cody'). The seeds were surface-sterilized by soaking in 5% chlorox solution (sodium hypochlorite) for 5 min before planting. For laboratory germination experiments, final test concentrations of mirex were prepared from a stock solution of recrystallized technical mirex dissolved in acetone and diluted with water. Twenty ml of acetone without mirex accordingly diluted with 4 liters of water, and water without acetone served as controls.

Germination experiments were conducted under constant temperature (25C). Four replicates of 50 seeds each for each species were planted on 15 x 30 cm germination blotters presoaked in the prepared mirex test solutions. After planting, the blotters were folded and placed in "Ziploc" plastic bags to keep them moist. During the germination period, tap water was used to keep the blotters moist. The plastic bags were placed in an automatic daylight controlled growth chamber. Germination counts were begun 4 days after planting and continued daily for 21 days on which day all the tests were terminated. Percent germination based on number of seeds planted was recorded.

Emergence and seedling growth experiments were conducted in a sandy substrate. Sand was used because it contains only a trace amount (0.04%) of organic matter which can bind and retain mirex residue. The other characteristics of the sand used and the manner by which mirex was applied to the test substrate to achieve different concentrations have already been described by us elsewhere in this volume. Sand treated with 100 ml acetone in 1 liter water and sand treated only with water served as controls.

The 5 kg sand for each test concentration was held in aluminum germination trays (50x30x10 cm). A total of 400 seeds of each crop was planted in 4 replicates consisting of 4 rows of 25 seeds for each replication. The seeds were covered with a thin layer of sand during planting. Five grams of 13-13-13 fertilizer per tray was applied in the initial irrigation.

Emergence counts were started 4 days after planting and continued daily up to 21 days, when no more seedlings emerged. Per cent emergence was based on number of seeds planted. A seedling was considered emerged when the coleoptile or hypocotyl emerged through the sand.

A duplicate emergence test similar to the above was conducted for seedling dry weight determination. All the emerged seedlings for each crop were clipped at the base immediately above the sand surface two weeks after planting and dried for 12 hrs at 120C. The dry weight obtained was computed on the basis of 100 seedlings.

RESULTS AND DISCUSSION

GLC analysis of the experimental planting substrate revealed final mirex concentrations of 0.15, 0.3, 0.7, 3.5, 7.5, 12.0, and 20.0 ppm. The results of the germination and emergence experiments shown in Table 1 indicate a significant (at 5% level) reduction in germination and emergence as the concentration of mirex increased. A close visual examination of seedlings revealed that some of the seedlings were poorly developed suggesting that the uptake of mirex by the young seedlings caused damage. The results also revealed that acetone, which is used as a "carrier" to bind mirex in the sand, had no effect on the emergence or germination of seeds.

The effects of mirex on seedling growth rate on the basis of dry weight of 100 seedlings after 2 weeks growth for each crop are presented in Table 2. It can be seen that seedling dry weight also decreased at higher amounts of mirex concentrations. Crimson clover, johnson grass, and annual rye grass showed initial significant reduction in growth rate at 0.15 ppm mirex concentration; tall fescue and alfalfa at 0.30 ppm concentration; and alsike clover at 0.70 ppm.

Insecticide and herbicide suppression of germination, emergence, survival and seedling growth of crop seeds have been demonstrated earlier by works of CULLINAN (1949), KAPPELMAN and BUCHANAN (1968), CHANG and FOY (1971), RANNEY and HEARTLEY (1972), PHILLIPS et al. (1972). The selective nature of mirex phytotoxicity of crop seeds observed by us agrees with the findings of CHANG and FOY (1971), GAWAAD et al. (1972b), and COX and LILLY (1952).

The exact mechanism and mode of toxicity of mirex on germination and development of seedlings has not yet been fully resolved. We have observed in one of our studies (elsewhere in this volume) that mirex is taken up and accumulated by seedlings of different crops. The uptake and accumulation of mirex was directly related to the mirex concentration in the rooting medium.

TABLE 1

Percentage germination and emergence of crop seeds exposed to mirex

Treatments	Crimson clover	Johnson grass	Annual Rye grass	Tall fescue	Alsike clover	Alfalfa
ols ter ter	93a * 92a	% Ge 57a 57a	% Germination a a 96a	86a 84a	87a 87a	83a 82a
0.30	84a 80b 71c	42ab 33b 30bc	90a 92 a 795	56b 49b 56b	63b 61b 53b	73b 67b 71ah
3.50 7.50 12.00 20.00	74c 71c 72c	33b 33b 28c 29c	78b 64c 68c 59c	36 c c s c c c c c c c c c c c c c c c c	555 500 450 500 500	69b 66b 62c 70b
C # #	94a 94a	% El 54a 54a	mergence 93a 92a	53a 53a	78a 78a	89a 88a
0.15 0.30 0.70	87a 80b 73c	40ab 30c 28c	86a 88a 74b	47a 36b 38b	58b 40c 38c	69b 69b 62b
3.50 7.50 12.00 20.00	65cd 67c 60d 51d	26c 23d 22d 23d	960 660 58cd 58d	32bc 30c 30c 28c	36c 30cd 22d 22d	66b 57c 58c 54c

* Means within an array followed by the same letter do not differ significantly at the 0.05 level according to Duncan's Multiple Range Test

TABLE 2

Dry weight (mg) of 100 seedlings of various crops after 2 weeks growth in sand contaminated with different concentrations of mirex.

Treatments	rs.	Crimson clover	Johnson grass	Annual Rye grass	Tall fescue	Alsike clover	Alfalfa
Controls Water	itrols Water alone	786a*	755a	684a	694a	656a	775a
Water	Water & acetone	780a	750a	687a	690a	640a	760a
Mirex	(mdd)						
	0.15	721b	637b	577b	675a	630a	750a
	0.30	700b	677b	250b	290b	600a	650b
	0.70	710b	q099	562b	618b	430b	484c
	3,50	536c	571c	387d	550b	398b	484c
	7.50	535c	548c	440c	413c	380c	394c
, -	12.00	558c	513d	428c	385c	332c	353c
V	20.00	575c	518d	412c	392c	304c	380c
			-				

* Means within an array followed by the same letter do not differ significantly at the 0.05 level according to Dunçan's Multiple Range Test.

The reduction in seed germination, seedling emergence and growth reported in the present study could be due to a rapid diffusion of water containing mirex into germinating seeds during the germination process.

The results of this study indicate that total germination, seedling emergence, and early growth are reduced in several plant species when exposed to concentrations of mirex exceeding 0.15 ppm. Normally the amount of mirex (incorporated in the bait) applied to areas infested by fire ants is small (4.2 g per hectare). No data are available to establish the exact quantity of this insecticide in the soil. Increasing concern about the effect of mirex on non-target organisms and its fate in the environment prompted the present investigation on the effect of mirex on germinating seeds and growing seedlings.

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