

Translocation of Four Organochlorine Compounds by Red Mangrove (*Rhizophora mangle* L.) Seedlings¹

by GERALD E. WALSH, TERRENCE A. HOLLISTER, and JERROLD FORESTER

*U.S. Environmental Protection Agency
Gulf Breeze Environmental Research Laboratory
Sabine Island, Gulf Breeze, Fla. 32561
(Associate Laboratory of the National Environmental
Research Center, Corvallis, Ore.)*

Mangrove vegetation is common in tropical estuaries and serves as a habitat and source of food for many animals (ODUM 1971). Because mangrove, both living and detrital, is eaten by a variety of animals, we conducted studies to learn translocational patterns of four organochlorine compounds in seedlings to determine if these persistent compounds could be introduced into estuarine food webs that receive contributions from mangrove.

Previous studies showed that mangroves and other plants translocated certain toxicants from the soil to leaves. Seedlings of the red mangrove (*Rhizophora mangle* L.) translocated the herbicides 2,4-D and picloram from soil to roots, hypocotyls, stems, and leaves (WALSH et al. 1973). Translocation of the insecticides dieldrin by alfalfa and hay (MUMMA et al. 1966), dieldrin, endrin, and heptachlor by soybeans (NASH and BEALL 1970), and mirex by peas and beans (MEHENDALE et al. 1972) has been demonstrated.

Insecticides have been found to be associated with mangrove from natural stands. Dieldrin (0.021 ppm - parts per million) and polychlorinated biphenyls (0.181 ppm) were found in red mangrove leaves from St. John and St. Croix in the Virgin Islands². We found DDD in roots (0.022 ppm), hypocotyls (0.220 ppm), stems (0.032 ppm), and leaves (0.019 ppm) of red mangrove seedlings from Joyuda, Puerto Rico (unpublished data).

In the present study, we investigated translocation of the insecticides dieldrin, methoxychlor, and mirex and the polychlorinated biphenyl (PCB) Aroclor® 1242 by red mangrove seedlings in the laboratory.

METHODS

Seedlings 18.5 to 38.2 cm long were obtained from trees in the Loxahatchee River near Jupiter, Florida, and planted in plastic boxes that contained muddy sand and natural sea water from an estuary near Gulf Breeze, Florida. Fifteen seedlings were planted in each box. Salinity of the water that covered the sediment was 25 parts per thousand; pH of the sediment was between 6.3 and 6.7. Air temperature was 20 - 23° C. Light was provided by overhead Grow-Lux® fluorescent tubes in a regime of alternate 12-hr periods of light and darkness.

¹ Contribution No. 193, Gulf Breeze Environmental Research Laboratory.

² Philip A. Butler (Gulf Breeze Environmental Research Laboratory, Gulf Breeze, Florida). Personal communication: Unpublished data, EPA National Monitoring Program.

Technical grade dieldrin (1,2,3,4,10,10-hexachloro-6,7-epoxy-1,4,4a,5,6,7,8,8a-octahydro-1,4-endo-exo-5,8-dimethanonaphthalene), methoxychlor (1,1,1-trichloro-2,2-bis (p-methoxyphenyl) ethane), mirex (dodecachlorooctahydro-1,3,4-metheno-1H-cyclobuta (cd) pentalene), and Aroclor 1242 (a mixture of polychlorinated biphenyl isomers), dissolved together in 10 ml of acetone, were added to the surface of the water. The same amount of acetone was added to eight control boxes. Application rates were 0.06, 0.11, 0.28, 0.56, 1.12, 2.80, 5.60, and 11.20 kg/ha (0.05, 0.10, 0.25, 0.50, 1.00, 2.50, 5.00, and 10.00 lb/acre). These rates were equal to concentrations of 0.038, 0.075, 0.150, 0.300, 0.600, 1.50, 3.00, and 6.00 ppm in the muddy sand.

Three boxes of seedlings were treated at each application rate after one or two pairs of leaves had emerged from the stems. Two or three seedlings were collected from each box (total of 8 or 9 plants per sample) each week for six weeks after application. The seedlings were washed with tap water and then with acetone. Roots, hypocotyls, stems, and leaves of plants from each concentration and the controls were analyzed separately for organochlorine residues.

Samples were homogenized in a blender with four times their weight of anhydrous sodium sulfate, then extracted for four hours with 10% ethyl ether in petroleum ether in a Soxhlet apparatus. The extract was concentrated to approximately 15 ml and transferred to a florisil column (MILLS et al. 1965).

Methoxychlor, mirex, and Aroclor 1242 were eluted with 6% ethyl ether in petroleum ether; dieldrin with 15% ethyl ether in petroleum ether. Approximately 1 ml of metallic mercury was added to approximately 5 ml of the extracts of roots and hypocotyls to remove sulfur compounds that interfered with electron capture gas chromatography.

The samples were analyzed with a model 2100 Varian Aerograph gas chromatograph equipped with 182.8 cm X 2 mm I.D. glass columns and electron capture detectors. Two columns were packed with 2% OV - 101, one with 3% OV - 210, and one with a 1:1 ratio of 2% OV - 101 and 3% OV - 210, all on Gas Chrom Q. Quantitation was made on the OV - 101 columns. The other columns were used to confirm the analyses. Carrier gas was pre-purified nitrogen. Operating conditions were: injector, 210° C; columns, 195° C; detectors, 215° C; gas flow, 25 ml/min.

Aroclor 1242 was quantified by measurement of total peak height of the 12 major peaks, which were compared with heights of the same peaks in a standard solution of known concentration. The other compounds were quantitated by peak height.

® Aroclor, Registered Trademark, Monsanto Co., St. Louis, Mo.; Gro-Lux, Sylvania Electric Products Inc., Salem, Mass. Mention of commercial products does not constitute endorsement by the Environmental Protection Agency.

Recovery rates were above 80% in quality-control samples to which known amounts of the compounds had been added. Residue data do not include a correction factor for recovery rate.

RESULTS AND DISCUSSION

Mangrove seedlings translocated the four organochlorine compounds tested, but no visible effects of the compounds on the seedlings were noted. None of the compounds was detected in control seedlings.

Dieldrin

Dieldrin was translocated to hypocotyls and leaves more rapidly than were the other compounds. It was detected in hypocotyls and leaves one week after exposure at all concentrations and these plant parts contained more of the chemical than did roots and stems (Table 1). Dieldrin was never detected in stems and was not found in roots at exposure concentrations less than 0.28 kg/ha. Accumulation of dieldrin in leaves was not related to length of exposure: concentrations were similar during the entire exposure. Average residues in leaves ranged from 0.072 to 0.113 ppm but were not related directly to exposure concentrations.

TABLE 1

Average concentrations of dieldrin in roots, hypocotyls, stems, and leaves of red mangrove seedlings during six weeks of exposure to eight application rates

<u>Application Rate</u> <u>kg/ha</u>	<u>Average Concentration, Parts Per Million</u>			
	<u>Roots</u>	<u>Hypocotyl</u>	<u>Stem</u>	<u>Leaves</u>
0.06	ND*	0.01	ND	0.07
0.11	ND	0.02	ND	0.11
0.28	Tr**	0.02	ND	0.11
0.56	Tr	0.04	ND	0.07
1.12	0.03	0.08	ND	0.11
2.80	0.04	0.08	ND	0.07
5.60	0.04	0.08	ND	0.09
11.20	0.06	0.16	ND	0.10

* Not detected; ** Trace, present but not quantifiable. Limit of detection = 0.01 ppm.

Concentrations of dieldrin in hypocotyls were often related directly to application rates and to length of exposure. At application rates of 1.12 kg/ha and above, concentrations in hypocotyls increased as exposure time increased (Figure 1). Concentrations of dieldrin detected in hypocotyls at the end of the six-week exposures were less than concentrations of methoxychlor, mirex, and Aroclor 1242 in hypocotyls of seedlings exposed at the same concentration rates.

Methoxychlor

Methoxychlor was present in roots at the three highest application rates and was translocated to hypocotyls of seedlings treated at rates of 0.28 kg/ha and above (Table 2). It was never detected in stems or leaves.

Residues in hypocotyls increased throughout the six-week exposure and were related directly to application rates above 0.56 kg/ha at the end of the exposure (Figure 2).

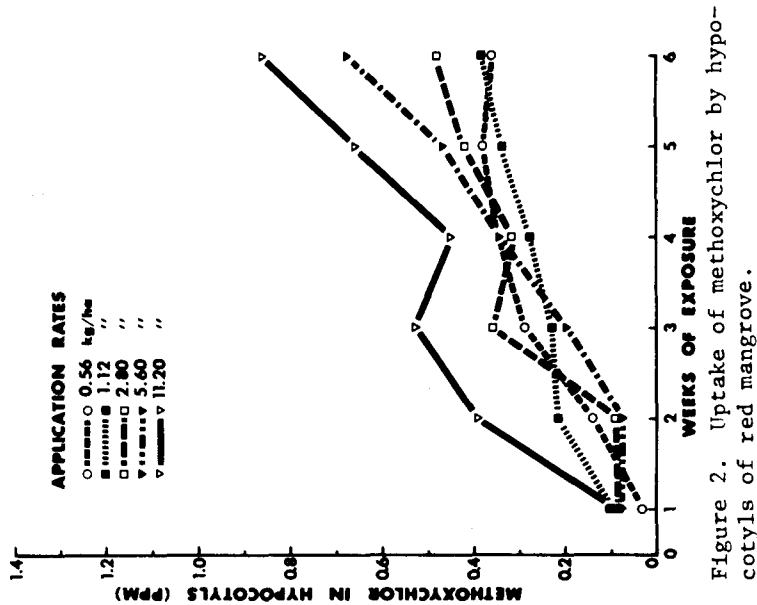


Figure 2. Uptake of methoxychlor by hypocotyls of red mangrove.

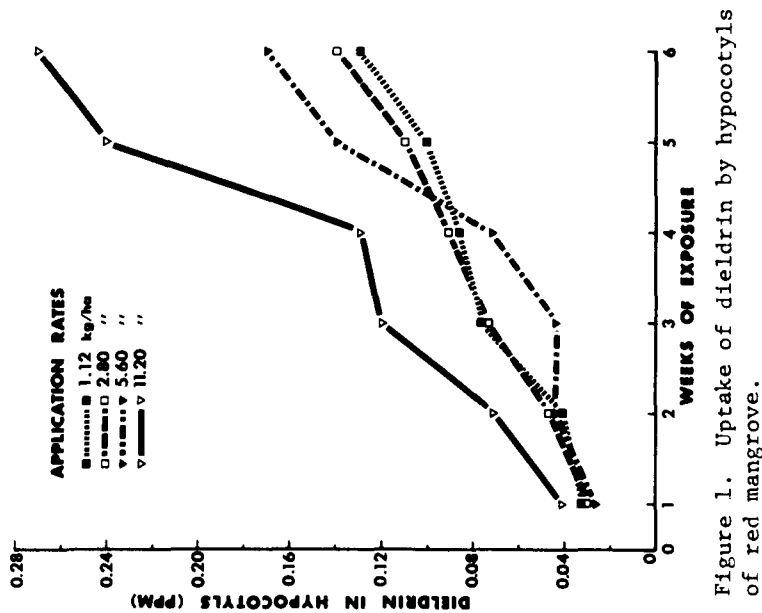


Figure 1. Uptake of dieldrin by hypocotyls of red mangrove.

TABLE 2

Average concentrations of methoxychlor in roots, hypocotyls, stems, and leaves of red mangrove seedlings during six weeks of exposure to eight application rates

<u>Application Rate</u> kg/ha	<u>Average Concentration, Parts Per Million</u>			
	<u>Roots</u>	<u>Hypocotyl</u>	<u>Stem</u>	<u>Leaves</u>
0.06	ND*	ND	ND	ND
0.11	ND	ND	ND	ND
0.28	ND	0.03	ND	ND
0.56	ND	0.26	ND	ND
1.12	ND	0.26	ND	ND
2.80	0.02	0.29	ND	ND
5.60	0.06	0.31	ND	ND
11.20	0.07	0.49	ND	ND

* Not detected. Limit of detection = 0.02 ppm.

Mirex

Mirex was detected in seedlings treated at 11.20 kg/ha but not at lower application rates (Figure 3). Residues were greatest in all plant parts after two weeks of exposure and decreased rapidly thereafter. It was not found in leaves after six weeks of exposure and only 0.03 ppm was detected in roots at that time.

Aroclor 1242

Aroclor 1242 was detected in roots of seedlings exposed at application rates of 5.60 and 11.20 kg/ha but not in stems. It

TABLE 3

Average concentrations of Aroclor 1242 in roots, hypocotyls, stems, and leaves of red mangrove seedlings during six weeks of exposure to eight application rates

<u>Application Rate</u> kg/ha	<u>Average Concentration, Parts Per Million</u>			
	<u>Roots</u>	<u>Hypocotyl</u>	<u>Stem</u>	<u>Leaves</u>
0.06	ND*	ND	ND	ND
0.11	ND	ND	ND	ND
0.28	ND	ND	ND	ND
0.56	ND	0.35	ND	0.46
1.12	ND	0.79	ND	1.11
2.80	ND	0.92	ND	0.92
5.60	0.10	0.53	ND	0.80
11.20	0.10	1.50	ND	0.90

* Not detected. Limit of detection = 0.1 ppm.

Fig. 3

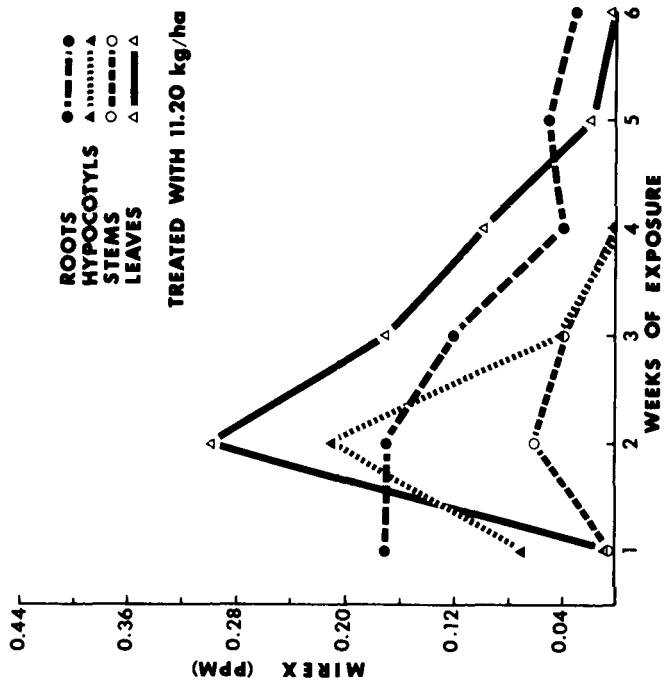


Figure 3. Mirex in roots, hypocotyls, stems and leaves of red mangrove treated with 11.20 kg/ha.

Fig. 4

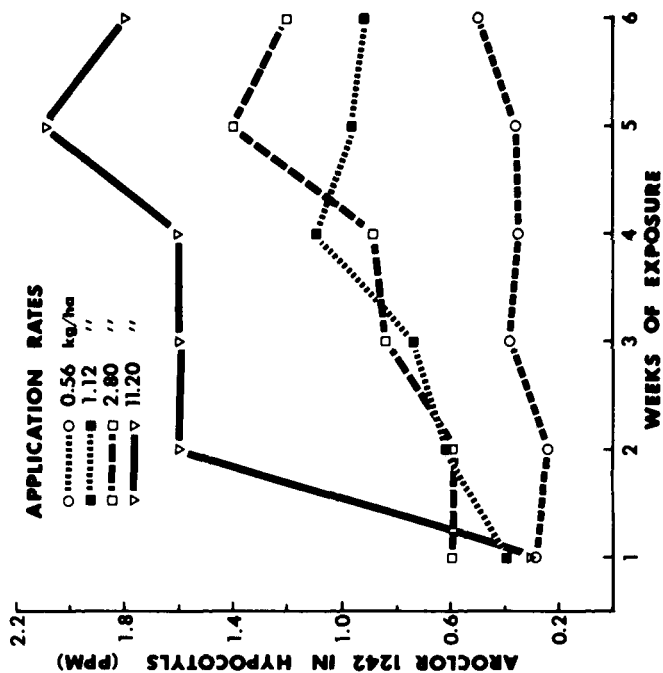


Figure 4. Uptake of Aroclor 1242 by hypocotyls of red mangrove.

was detected in hypocotyls and leaves at application rates of 0.56 kg/ha and greater (Table 3).

Concentrations of Aroclor 1242 in leaves did not change with time at any treatment rate, but concentrations in hypocotyls increased in relation to exposure time for at least five weeks (Figure 4). In hypocotyls, concentrations of PCB were greater than concentrations of each of the other three compounds.

SUMMARY

Mangrove seedlings from the field were found to contain DDD, dieldrin, and PCBs.

In the laboratory, mangrove seedlings translocated dieldrin, methoxychlor, mirex, and Aroclor 1242 (a PCB) from soil to various plant parts. Dieldrin was detected in hypocotyls and leaves of seedlings exposed to application rates of 0.06 kg/ha and above; methoxychlor in hypocotyls at rates of 0.28 kg/ha and above; Aroclor 1242 in hypocotyls and leaves at rates of 0.56 kg/ha and above; and mirex in roots, hypocotyls, stems, and leaves only at the highest treatment rate of 11.20 kg/ha.

The data show that these persistent organochlorine compounds can be translocated to seedlings. If the compounds are present in the natural mangrove environment, it is possible that they could enter seedlings and pass to higher trophic levels when seedlings are eaten by estuarine organisms.

ACKNOWLEDGEMENTS

We thank Mr. Robert L. Goodrick of the Central and Southern Florida Flood Control District for providing the seedlings used in these studies. We also thank Dr. Philip A. Butler, EPA, Gulf Breeze Environmental Research Laboratory, for residue data on mangroves from the Virgin Islands, and Dr. Seppo E. Kolehmainen, Puerto Rico Nuclear Center, for sending seedlings from Puerto Rico.

REFERENCES

- MEHENDALE, H. M., L. FISHBEIN, M. FIELDS, and H. B. MATTHEWS. Bull. Environ. Contam. Toxicol. 8, 200 (1972).
- MILLS, P. A., J. F. ONLEY, and R. A. GAITHER. J. Assoc. Off. Agric. Chem. 46, 182 (1965).
- MUMMA, R. O., W. B. WHEELER, D. E. H. FREAR, and R. H. HAMILTON. Science 152, 530 (1966).
- NASH, R. G. and M. L. BEALL, JR. Science 168, 1109 (1970).
- ODUM, W. E. Univ. Miami, Sea Grant Tech. Bull. No. 7, 162 p. (1971).
- WALSH, G. E., R. BARRETT, G. H. COOK, and T. A. HOLLISTER. Bio-science 23, 361 (1973).