Mirex Residues in Wild Populations of the Edible Red Crawfish (*Procambarus clarki*)

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The rearing of the red crawfish, <u>Procambarus</u> <u>clarki</u> (Girard), in rice fields and ponds or trapping in canals, bayous, and swamps for sale in local markets is a multimillion-dollar-a-year industry in Louisiana(1).

Crawfish, like many other crustaceans, are sensitive to many of the chlorinated hydrocarbon insecticides including DDT (2,3,4,5), dieldrin, and endrin (6). Aldrin has also been reported as being moderately toxic to crawfish in the laboratory (7), but at least one field study revealed no effect on survival, growth, or reproduction when aldrin was applied at the recommended treatment rate of 0.25 or 0.5 lbs per acre as a seed dressing (8).

Mirex is basically related to the other chlorinated hydrocarbon insecticides, although it is totally chlorinated. The earliest studies of mirex (2) showed adult crawfish, Procambarus clarki, to be unaffected by up to 3 days' exposure to 68 times the recommended rate of mirex. A more recent study (9) has reported that juvenile forms of two related crawfish (Procambarus blandingi, and P. hayi) were extremely susceptible to mirex bait, or exposure to mirex dissolved in water, and were capable of concentrating mirex 27,210 fold from the water in which they were held.

Mirex is widely used for controlling the imported fire ant, <u>Solenopsis</u> <u>saevissima</u> <u>richteri</u> Forel, an agricultural and human pest in Louisiana. Mirex bait is retailed to homeowners in 5 pound bags containing .15% actual toxicant.

A second formulation containing .3% mirex is furnished to farmers at cost through a State extension program. In 1970, in excess of 280,000 pounds of bait were distributed through this farmer program. In north Louisiana, outside the crawfish growing area, Federal/State cooperative programs have treated large blocks of land to retard the spread of the fire ant into adjacent states of Arkansas and Texas. In 1971, 18,750 pounds of bait were used in this program.

In light of the laboratory findings of sensitivity of crawfish to mirex and the amount used in crawfish-growing areas of Louisiana, it would appear that the continued use of this pesticide could threaten this industry. To evaluate the problem we

have undertaken this study to determine if mirex residues in crawfish from treated areas are beginning to approach those levels found under laboratory conditions to affect crawfish (9). Secondarily, we felt it would be useful to determine if unexpectedly high mortality or population declines have been observed by farmers who commercially raise crawfish following the use of mirex around their ponds.

MATERIALS AND METHODS

Sample Collection:

Collections of samples were made in May 1971 during the peak of the crawfish harvests. The area chosen for the study was in southcentral Louisiana and is generally considered to be the center of the crawfish-producing region. Five USDA, PPQP inspectors stationed in this area and basically familiar with the crawfish industry were assigned the job of collecting the specimens and interviewing the growers.

A portion of the commercial crawfish crop came from rice fields where they are raised during winter as an alternate crop. Originally we had planned to do extensive sampling in rice fields but learned that the nature of rice farming, flooding and extensive use of other pesticides, normally prevents heavy infestations of the imported fire ant from becoming established. Consequently, comparatively little mirex is used in rice fields, although some is used to treat the levees surrounding the fields.

All collections, therefore, were made from ponds, canals, bayous, etc., which had a history of mirex bait used in pastures immediately adjacent to the water. All sites, however, were being used to raise crawfish (1). The inspectors were instructed to obtain an 0.5 kg sample (20 to 35 live crawfish) from the grower at the time of harvest and interview him to obtain his observations on the abundance of crawfish over the past few years. Since the production of crawfish often represents a major source of income for many of these people, their observations were felt to be basically valid.

The live crawfish samples were cooled until immobile, wrapped in several layers of aluminum foil, placed in plastic bags along with an identification number, frozen, and shipped to the APHIS chemical laboratory in Gulfport, Mississippi.

Sample Processing:

Samples were washed in tap water to remove mud, algae, etc., and the entire sample ground and mixed in a Waring blender. A 50-gram subsample was removed and mixed with 300 ml nanograde hexane and 100 ml isopropyl alcohol, and rotated for 2 hours. The extract was filtered through prewashed glass wool and divided into two equal subsamples, one to be used for analysis and the other preserved as a check. The portion to be analyzed was washed three times in distilled water, filtered through

anhydrous NaSO4 and glass wool, and evaporated to the desired volume. The extract was treated with concentrated sulfuric acid to clean up the background, rewashed and filtered again with anhydrous NaSO4 and finally cleaned up with a florisil column. The final sample, plus the hexane washes, was concentrated to 2.5 ml in a water bath with an air stream.

Sample Analysis:

Primary analysis was done using a Microtek Model 220 equipped with an electron capture detector. The column (6' x 1/2") was packed with 1.5% OV-17 and 1.96 QF-1 on Gas Chrom Q. Retention time was 30 minutes with 80 ml gas flow (5% Argon-95% Methane) with injector, oven, and detector temperatures of 250°, 200°, and 210°C respectively. Confirmation of all samples was done on Hewlett Packard Model 402 using a column (6' x 1/4") packed with 3% DC-200 on Gas Chrom Q. With a flow rate of 100 ml/min (5% Argon-95% Methane) and the injector, oven, detector temperatures of 245°, 175°, and 205°C respectively, retention time was 36 minutes. Recovery of mirex averaged 75-80% when fortified samples were taken through the complete process.

Samples were also analyzed for DDT and its byproducts (TDE, DDE), chlordane, toxaphene, and the PCB arochlor 1260. Level of detection by this method was 0.01 ppm. Dieldrin, aldrin, endrin cannot be detected using this technique since they have been destroyed during the sulfuric acid wash.

RESULTS AND DISCUSSION

The results of interviews and chemical analyses for mirex are presented in Table 1. Twenty-six of the samples collected came from farm ponds and large drainage ditches in pastures where mirex bait had been used. The inspectors doing the collection frequently found that the water itself had been treated when an application of mirex was made by small aircraft or when the owner tried to reach mounds on dam, levees, or spoil piles beside canals. Also, most areas were found to have been treated with at least twice the recommended rate of 1 1/4 lbs bait per acre (1.7 grams technical mirex per acre), due to the difficulty in calibrating most ground spreader equipment. Swamps and bayous immediately adjacent to treated areas were also used for three of the collection sites. No samples were taken from the Atchafalaya Spillway, a huge swamp on the western side of the Mississippi River, which is a major source of crawfish, but which has no history of treatment by mirex.

Of the locations chosen for sampling, 13 had been treated only once. The remainder had received yearly treatments for up to 7 years. Seven of the samples did show detectable residues of mirex, but these residues could neither be associated with frequency of the treatment nor the type of habitat from which the crawfish came. It was noted that the few residues detected were below the 1 to 8 ppm level reported killing juvenile crawfish in the laboratory (9). In the three locations where populations were reported to have below normal, none of the crawfish showed detectable mirex residues.

TABLE 1.

Crawfish collected in 7 parishes of southcentral Louisiana in May 1971. Level of detection for mirex and DDT 0.01 ppm. N. D. means none detected at this level.

		No. of				
		Times		Pestici	de Resi	Pesticide Residues - PPM
Source	Last Treated	Treated	Abundance	:Mirex	DOT	Other:
•						
St. Landry Parish						
Commercial pond	Spring '70	2	plentiful	Ø	. 22	PCB?
Commercial pond	Spring '70	2	plentiful	æ	. 22	PCB?
Commercial pond	June '70		below normal	Q	.12	PCB1
Commercial pond	Spring '70	-1	not plentiful	2	60.	
Drainage canal	Apr11 '70		no comment	Ø	.02	
Farm pond	June '69		normal	.01	80.	
Road ditch	Summer '69	-	no comment	QN	.37	
Farm pond	Summer '70	Ŋ	normal	Ø	80.	
Drainage canal	June '70	2	normal	Ø	.07	
Pond	June '69	-1	normal	.01	.13	.11 chlordane
Lafayette Parish						
Canal	Summer '70	2	no comment	QN	.23	
Canal	Summer '70	2	normal	Ø	90.	
Swamp	Spring '70	2	plentiful	ON	.05	
Evangeline Parish	1					
Pond	June '70	~ 4	normal	.01	.13	
Pond	Summer '70	2	plentiful	Ø	. 29	
St. Martin Parish			•			
Bayou	Summer '70	4	plentiful	<u>R</u>	. 20	PCB
Drainage canal	Summer '70	m	plentiful	QN N	. 28	PCB
Commercial pond	June '70	-	plentiful	.07	.07	
Pond	Spring '70	-	poor	Q	.08	

		No. of					
		Times		: Pestic	ide Res	:Pesticide Residues - PPM	••
annoc	Last Treated	Treated	Abundance	:Mirex DDT	DDT	Other	
St. Martin Parish (con't)	(con't)						
Drainage canal		٣	no comment	Ę	20		
Commercial pond		_	plentiful	2 5	3 3		
Commercial pond	Spring '70	2	plentiful	3 5			
Avoyelles Parish			1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1	•	7		
Swamp	May '70	4	very abundant	70	3,0		
Pond	May-August '70	7	plentiful	Ę	? *		
Canal	August '70	2	normal	2 5			
Canal	April '71	Ŋ	normal	2 5	07.		
Canal	April '71	7	above normal	2	٠, ۵		
Vermillion Parish	1			2	3		
Swamp	August '70	5	very abundant	QX	90.		

Even if a significant population decline or kill had been reported, it would have been difficult to directly correlate it to the use of mirex. High levels of DDT and its byproducts were found in all crawfish sampled. Since it is known that crawfish are very sensitive to DDT (2,3,4,5), this would appear to be a more likely source of mortality than mirex. Similarly, residues of other pesticides have been reported in soil and water of crawfish-growing areas (10) and would also have to be monitored for and evaluated to determine their role in any reported kills. Finally, the biology of the crawfish must be kept in mind when evaluating reported kills, since a natural die-off of adults occurs in the summer and late fall after they have reproduced.

SUMMARY

Despite laboratory evidence of the extreme sensitivity of crawfish to the insecticide mirex, field studies and monitoring reveal no evidence of massive die-off or noticeable population declines. The residues obtained were so low as to be well below the levels capable of causing mortality of juvenile crawfish in the laboratory. It was concluded, therefore, that the present use of mirex in southcentral Louisiana is not an important threat to the crawfish industry.

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