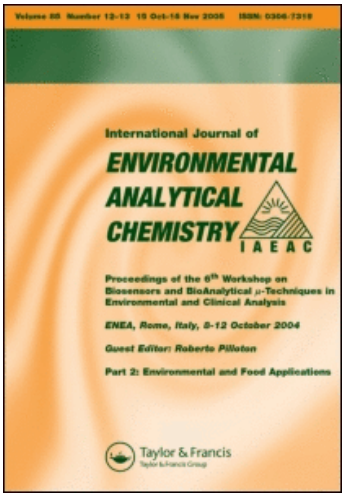


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S. Calero[a]; I. Fomsgaard[ab]; M. L. Lacayo[a]; V. Martinez[a]; R. Rugama[a]
[a] Research Center for Aquatic Resources of Nicaragua, Managua, Nicaragua [b] Department of Pesticides and Ecotoxicology, Research Centre for Plant Protection, Slagelse, Denmark

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# TOXAPHENE AND OTHER ORGANOCHLORINE PESTICIDES IN FISH AND SEDIMENT FROM LAKE XOLOTLÁN, NICARAGUA

S. CALERO, I. FOMSGAARD,[§] M.L. LACAYO, V. MARTINEZ and R. RUGAMA

*Research Center for Aquatic Resources of Nicaragua, PO Box 4598, Managua, Nicaragua*

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The levels of 11 organochlorine pesticides were analyzed in samples of two fish species (*Sarotherodon mossambicus* and *Cichlasoma managüense*) and sediments. Due to a toxaphene-producing factory located on the shore of Lake Xolotlán, toxaphene was detected in more than 80% of the fish specimens and in all the sediment samples analyzed. α-BHC, heptachlor, heptachlor-epoxide, aldrin and dieldrin were detected neither in fish nor in sediment samples. DDT or its metabolites DDE or DDD (ΣDDT) were present in almost all the fish and sediment samples but in low concentrations. The presence of β-BHC and lindane (τ-BHC) in fish and lindane in sediment was insignificant.

**KEY WORDS:** Organochlorines, toxaphene, fish, sediment, tropical lake, Nicaragua.

## INTRODUCTION

Organochlorine compounds, such as DDT, the drins, BHC's and toxaphene, are environmentally persistent compounds which have a capacity for bioaccumulation and biomagnification and produce toxicity in aquatic organisms which are consumed by the population.[1]

Toxaphene is also known as chlorinated camphene, Synthetic 3956, polychlorocamphene, camphechlor, Chlor Chem-T-590, Cristoxo, Moto, Phenacide, Phenatox, Strobane-T, Toxon 63 and Vapotone.[2] It is an insecticide used to combat caterpillars, grasshoppers, the cotton boll weevil and ectoparasites on cattle and sheep.[3]

In Nicaragua, the factory HERCULES of Central America has been producing toxaphene since 1974, when it produced 9,500 metric tons of the pesticide. The factory is located on

[§] Present address: Research Centre for Plant Protection, Department of Pesticides and Ecotoxicology, Flakkebjerg, DK-4200 Slagelse, Denmark.

the outskirts of the city of Managua, on the shore of Lake Xolotlán, and it discharges all its waste products into this ecosystem. In the year 1976 the factory produced 5,700 metric tons of toxaphene. Since that year the production has been varying, with a tendency to decline; in 1985 the production was 2,140 metric tons and in the year 1989 it was 300 metric tons. From the year 1985 on, all the production has been sold in Nicaragua.[4,5] In 1991 the factory was working very irregularly, and since June 1991 it has been out of production.[5]

In developing countries such as Nicaragua, organochlorine pesticides like DDT are still used in vector control programs.[5] According to Matus and Beck,[5] the use of toxaphene has been restricted by law for the cultivation of cotton only, where it has been employed in great quantities. It is not known if it has been or still is used in other types of cultivation.

During the last five decades, Lake Xolotlán has been used as the recipient of domestic and industrial wastewater from the city of Managua. In addition, Lake Xolotlán receives the superficial runoff from its extensive drainage basin which is intensively cultivated.

The aim of the present study was to examine the concentration of 11 organochlorine compounds in fish and sediment from Lake Xolotlán. No other investigation on organochlorine pesticide residues in a Nicaraguan ecosystem has been published so far.



## MATERIALS AND METHODS

### *Fish*

Samples of the most abundant species in Lake Xolotlán, *Sarotherodon mossambicus* (tilapia) (56 specimens) and *Cichlasoma managüense* (guapote) (52 specimens) were collected in March 1991 from local fishermen at four sampling stations (Figure 1.) The samples were stored at -18°C until analysis.

A 10 g piece of the dorse lateral muscle from each fish were homogenized in acetone/n-hexane, and later extracted in two portions from a mixture of n-hexane/diethyl ether 9:1. The combined liquid phases were agitated with a solution of NaCl and $H_3PO_4$. The organic extract was evaporated with a light nitrogen pressure in a water bath at 60°C, and the residue was weighed to calculate the fat content. The fat was later diluted in 10 ml of n-hexane.[7] For the removal of the fat and the confirmation of the presence of the compounds to be examined, three types of treatments, acid, alkaline and nitration, were carried out. Acid treatment consisted of applying concentrated $H_2SO_4$ to an aliquot of the extract.[7] In the alkaline treatment, an aliquot of the extract was treated with a solution of alcoholic potassium hydroxide and sodium chloride in $H_3PO_4$.[7] In the case of nitration, an aliquot of the extract was evaporated to dryness and a mixture of strong acids $HNO_3$-$H_2SO_4$ was added. After the treatment with acids, the nitrous compounds were reduced with iron.[8] The reaction of the compounds appears in Table 1. All the final extracts were brought up with internal standard HCB to a concentration of 20 ng/ml. This fungicide has never been used in Nicaragua, and samples analyzed without adding HCB showed never to contain this substance. The percentage of recovery exceeded 74% in all the compounds analyzed.

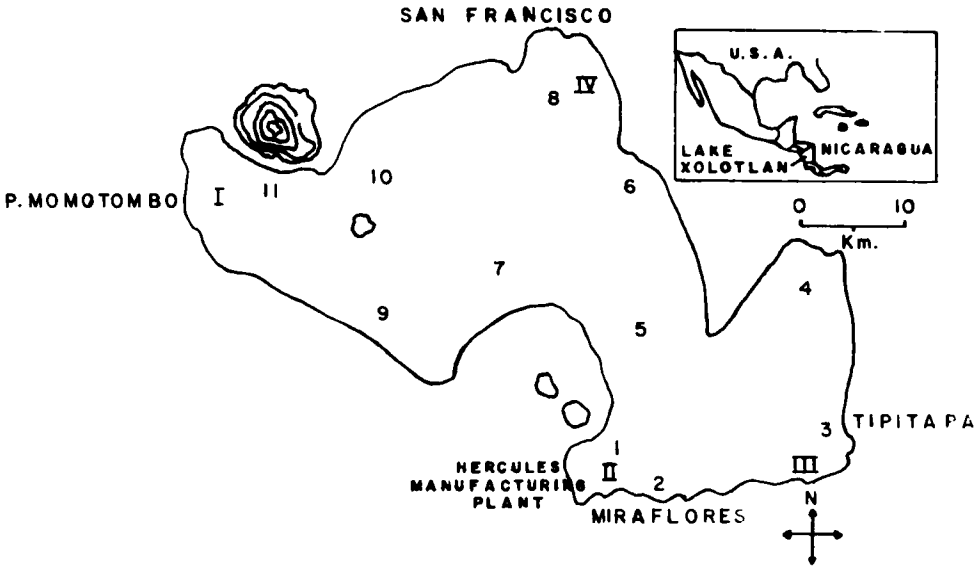


**Figure 1** Sample stations for fish and sediment samples in Lake Xolotlán, Nicaragua.

*Sediment*

Sediment samples were collected in the month of November 1991 at eleven sampling stations (Figure 1.) with an Ekman dredge (0–15 cm). The samples were preserved with acetone and stored in a refrigerator at 4°C until the time of analysis. Ten grams of centrifuged wet sediment were analyzed according to Jensen et al.[12] Elemental sulfur ($S_8$) generally present in sediments causes significant problems in analysis of residues. If the sulfur level is high the electronic capture detector becomes saturated for a considerable time. In order

**Table 1** Compounds analyzed and their reactions to the three types of applied treatments.

| | *Acid destr.* | *Alk. destr.* | *Nitration* |
|---|---|---|---|
| α-BHC | + | - | + |
| β-BHC | + | - | + |
| Lindane | + | - | + |
| Heptachlor | + | + | - |
| Aldrin | + | + | - |
| Heptachl. epox. | + | + | + |
| Dieldrin | - | + | - |
| p,p-DDE | + | + | - |
| p,p-DDD | + | - | - |
| p,p-DDT | + | - | - |
| Toxaphene | + | changes | + |

+ = resists
- = is eliminated

to eliminate this interference the sulfur was destroyed with tetrabutylammonium sulfite.[12] The acid, alkaline and nitration treatments were carried out in the same way as described for the fish. HCB 20 ng/ml was used as internal standard. The recoveries in the sediments exceeded 68% for all the compounds analyzed.

*Gas chromatographic determination*

A VARIAN 3400 gas chromatograph equipped with an electronic capture detector ($^{63}Ni$) was utilized for the identification and quantification of the compounds being studied. Three μl were injected in the splitless mode into a 30 m DB5 capillary column, with an internal diameter of 0.32 mm, using hydrogen as carrier gas with a linear velocity of 55 cm/s and nitrogen as make-up gas. The temperature program used was 80°C (1 min), 4°C/min 200°C, 3°C/min to 230°C, 15°C/min to 250°C (5 min). The temperature of the detector and of the injector were 320°C and 170°C.

Peaks were identified by comparing retention times with those of analytical standards obtained from Supelco Inc. The concentrations of the chlorinated pesticides were calculated on basis of a calibration curve using internal standard. All the compounds analyzed except dieldrin, p, p'-DDE and toxaphene were quantified directly from the acid destruction. The p, p'-DDE overlapped one of the major toxaphene peaks in the acid destruction. With nitration, p, p'-DDE is eliminated while toxaphene resists this treatment. Therefore by subtracting the p, p'-DDE apparent concentrations in these two methods, the correct value of p, p'-DDE was obtained. The analysis of toxaphene was problematic due to the complexity of the chromatogram, interference by other chlorinated hydrocarbons, and the differences in the chromatographic pattern between the analytical standard and weathered samples. Other authors have reported the same problems.[9,10,11] Applying the alkaline treatment both to the weathered samples and the toxaphene standard, very similar chromatograms of dehydrochlorinated toxaphene were achieved (Figure 2), with which toxaphene was quantified. Crist et al[9] quantified one major peak of toxaphene after alkaline dehydrochlorination. We added together the areas of the peaks 1 to 24 in the chromatogram (Figure 2) excluding

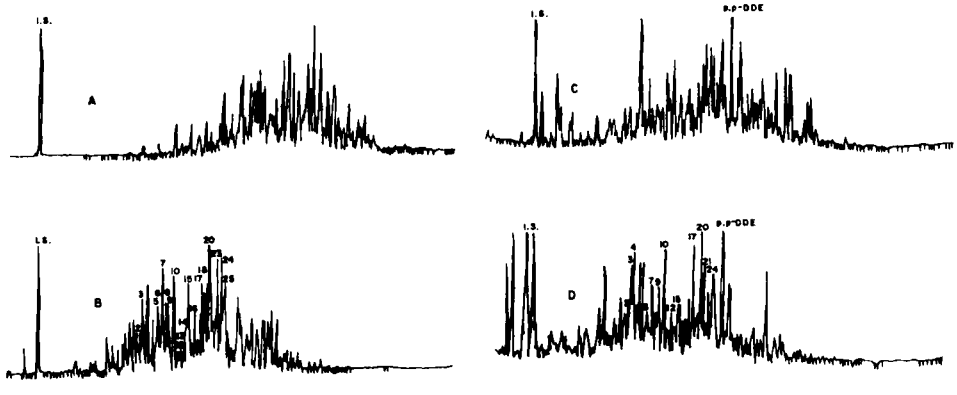


**Figure 2** A) Untreated toxaphene standard (Supelco). B) Toxaphene standard after alkaline treatment. C) Weathered sample. D) Weathered sample after alkaline treatment.

the p, p'-DDE peak which always was present after the alkaline treatment. According to Crist et al[9], the dehydrochlorinated quantitation is not possible in biological samples, when there are extensive metabolic or other structural changes. Such extensive changes were not observed in our investigation. The variations between the peak sizes are quite small, so we considered this quantitation method to be the best, when there is no acces to use a mass spectrometer. The only compound that could interfere in the selected retention time for the group of peaks was dieldrin. With the nitration it was confirmed that this compound was not present in any of the samples.

In all the samples the following compounds were analyzed: α-BHC, β-BHC, τ-BHC (lindane), heptachlor, heptachlor-epoxide, aldrin, dieldrin, p, p'-DDE, p,p'-DDD, p,p'-DDT and toxaphene. Both in fish and sediments the minimum detection limit for α-BHC was 0.5 $ng.g^{-1}$; for lindane, aldrin, heptachlor-epoxide and p, p'-DDE 1.0 $ng.g^{-1}$; for β-BHC, heptachlor, dieldrin, p, p'-DDD and p, p'-DDT 2.0 $ng.g^{-1}$; and for toxaphene (as the dehydrodechlorination products) 20 $ng.g^{-1}$. All figures expressed on wet weight basis.

## RESULTS AND DISCUSSION

### *Fish*

The results presented in Table 2 show the concentrations of organochlorine pesticides found in Lake Xolotlán in the two species of fish *C. managüense* (guapote) and *S. mossambicus* (tilapia). *C. managüense* is carnivorous and *S. mossambicus* is omnivorous.

The pesticides α-BHC, heptachlor, heptachlor-epoxide, aldrin and dieldrin were not detected in any of the fish analyzed from Lake Xolotlán.

The highest average concentration of toxaphene in the two species from the four stations of Lake Xolotlán was found in muscles of tilapia from Miraflores Bay, with 303 ng/g, and the lowest average concentration was 40 ng/g in tilapia from Momotombo.

In a study carried out by Mayer and Mehrle[13] it was found that fish of the species *Salvelinus fontinalis* with concentrations of 400 ng/g of toxaphene in muscle tissue showed reductions in growth, abnormal development of bones and reduced fertility. In Miraflores Bay the concentration in muscle of three specimens of *S. mossambicus* was higher than 400 ng/g; these fish could present similar symptoms although no investigation concerning that has been carried out so far.

On comparing the tilapias of Miraflores Bay with guapotes from the same place, where the toxaphene-producing factory is located, the tilapias contain concentrations of toxaphene 4–5 times higher than the guapotes both on fresh and fat weight basis. Therefore the variation between the two species could be due to differences in migratory habits.

In the year 1985 the Institute of Natural Resources and of the Environment[14] carried out a study of muscle tissue of *C. managüense* in Lake Xolotlán, and found a toxaphene concentration of 13.6 $\mu g.g^{-1}$ in the vicinity of the HERCULES factory. Klein[4] in 1987 detected a toxaphene concentration between 30 ng/g and 200 ng/g in muscle tissue in *C. managüense* from Lake Xolotlán; these dates are very similar to ours. The contrast between

**Table 2** Average concentrations and ranges of organochlorine pesticides in fish muscle tissue from Lake Xolotlán, Nicaragua (ng/g wet weight).

| *Station* | *Species* | *N* | *Wt. g*[♣] | *Fat %* | *β-BHC* | *Lindane* | *ΣDDT** | *Toxaphene* |
|---|---|---|---|---|---|---|---|---|
| I Momotombo | *Cichlasoma managüense* | 19 | 545 | 0.91 | 3 (8)[♦] (nd - 4) | 2 (6) (nd - 4) | 21 (11) (7 - 47) | 51 (8) (nd - 109) |
| | *Sarotherodon mossambicus* | 14 | 587 | 0.87 | 4 (13) (nd - 5) | 1 (7) (nd - 1) | 8 (13) (nd - 27) | 40 (14) (25 - 79) |
| II Bahia Miraflores | *Cichlasoma managüense* | 9 | 638 | 0.73 | nd | nd | 25 (9) (7 - 52) | 70 (9) (28 - 175) |
| | *Sarotherodon mossambicus* | 13 | 557 | 1.01 | 4 (10) (nd - 10) | 3 (1) (nd - 3) | 21 (13) (4 - 46) | 303 (13) (576 - 1131) |
| III Tipitapa | *Cichlasoma managüense* [§] | | | | | | | |
| | *Sarotherodon mossambicus* | 13 | 661 | 1.39 | 5 (4) (nd - 6) | 4 (3) (nd - 7) | 19 (11) (nd - 47) | 107 (8) (nd - 217) |
| IV San Francisco | *Cichlasoma managüense* | 24 | 786 | 1.59 | 5 (6) (nd - 8) | 2 (12) (nd - 3) | 30 (20) (9 - 114) | 79.2 (20) (24 - 171 |
| | *Sarotherodon mossambicus* | 16 | 658 | 1.67 | 4 (2) (nd - 5) | nd | 11 (14) (nd - 26) | 105 (16) (33 - 238) |

* ΣDDT = p,p-DDE, p,p-DDD and p,p-DDT
[♦] The number in parentheses indicates the number of positive values
[§] This species could not be caught during the time of sampling
[♣] Average values
N Number of samples
nd not detected

the results from 1985 and Klein's and our results must be due to the lowering of production in those years.

Examples of toxaphene residues in fish muscle tissue from different places around the world are those from the Great Lakes of the United States, with concentrations on fresh weight basis of 110 g/g in the years 1970–1974, 3,430 ng/g in the year 1977 and 2,370 ng/g in 1979.[15] Van der Valk y Wester[16] carried out a study in fish from northern Europe and found values from 1 ng/g in plaice filet to 67 ng/g in filet of herring. In Sweden where only 5,600 kg of toxaphene have been used, all before 1956, Andersson et al[17] analyzed fish from different ecosystems. On the west coast of Sweden they found 99 ng/g toxaphene in mackerel, and in salmon from Lake Vänern 213 ng/g. The authors suggest atmospheric transport as the cause, due to the relatively high vapor pressure of toxaphene.

Data on pesticide residues are frequently reported on a lipid weight basis, owing to the wide variety of sample matrices and their correspondingly diverse levels of lipid content. The tilapias of Miraflores Bay have an average toxaphene concentration of 30 μg/g fat and the tilapias of Momotombo 4.8 μg/g fat. These values are high compared to other values in the literature. Jansson et al[18] reported 13 μg/g toxaphene in muscle fat of herring (*Clupea harengus*) from the Baltic Sea. Arctic char (*Salvelinus fontinalis*) had 9 μg/g in muscle fat and alpine char (*Salvelinus alpinus*) 124 ng/g.[19]

Reimold and Durant[20] analyzed toxaphene residues in the aquatic flora and fauna in an estuary located close to a toxaphene-producing plant. The maximum value found in finfish was >200 μg/g fat. Data do not exist in the literature about the metabolism and period of excretion of toxaphene in aquatic organisms from tropical climates. The high medium temperature, 28.4°C, high alkalinity, 9.2, the high total bacterial concentration[21] and the strong ultraviolet radiation in Lake Xolotlán could accelerate the metabolism of toxaphene, These are probably some of the reasons that our values are not as high as those found by Reimold and Durant.[20] Another cause might be the low production of the toxaphene factory during 1991 in combination with a high capacity of fish to excrete toxaphene. Mayer et al[22] and Lowe et al[23] showed that fish eliminate toxaphene within a certain time if they are transferred to clean water.

The concentrations of ΣDDT in the fish from Lake Xolotlán are relatively low in comparison with the Baltic Sea, where Jensen et al[7] reported an average ΣDDT concentration in muscle tissue of 606 ng/g; according to the authors, this very high concentration probably is due to various factors. The low biomass, the low temperature and the low degree of interchange between this ecosystem and other waters are factors which influence the half life of DDT and its metabolites. In Lake Xolotlán the factors which may have influenced the rapid transformation of DDT are the same as those which could have affected toxaphene. In two Mexican lakes, Rosales and Escalona[24] found an average ΣDDT concentration of 3.13 ng/g in fish muscle on fresh weight basis.



### *Sediments*

The residues of organochlorine pesticides in sediments are presented in Table 3.

Neither α-BHC, β-BHC, heptachlor, heptachlor-epoxide, aldrin nor dieldrin were detected at any of the sampling stations.

**Table 3** Concentrations of organochlorine pesticides in sediment of Lake Xolotlán, Nicaragua (ng/g wet weight).

| *Station* | *Lindane* | *ΣDDT** | *Toxaphene* |
|---|---|---|---|
| 1 Hércules 100m disch. | n.d. | 102 | 175 |
| 2 Rubén Darío | 2 | 23 | 151 |
| 3 Tipitapa | 2 | n.d. | 62 |
| 4 Ba. San Antonio | n.d. | n.d. | 65 |
| 5 Pta. Huete | n.d. | 3 | 83 |
| 6 Pta. San Roque | n.d. | 3 | 116 |
| 7 Center | 3 | 2 | lost |
| 8 San Francisco | n.d. | 6 | 180 |
| 9 Mateare | n.d. | n.d. | 187 |
| 10 Isla Rosa | n.d. | 8 | 183 |
| 11 Geothermic plant | n.d. | n.d. | 62 |

*ΣDDT = p,p-DDT, p,p-DDD and p,p-DDT
n.d. = not detected

The toxaphene concentration in sediment from Lake Xolotlán was in a range of 63 – 187 ng/g wet weight. If the factory had been discharged during the time of sampling (november 1991) highervalues would probably have been found at sample point #1 (HERCULES, 100 m from discharge). In a former work[25] at the beginning of 1991, we detected 359 μg/g in sediment from the very discharge channel of the HERCULES factory, but we did not find toxaphene in samples of sediment from the recipient (Lake Xolotlán). The detection limit was 200 ng/g. With the knowledge that production in former years was higher, we suppose that the concentrations in the sediment were also higher, but there are no reports with regard to this.

In a study carried out in an estuary in Georgia, USA, which received residual water from a HERCULES INC. toxaphene-producing factory, Durant and Reimold[26] and Reimold and Durant[20] found toxaphene concentrations in surface sediments of 35.5 up to 1,858 μg/g.

Lindane was only detected at two sample stations and the concentration was relatively low. At station #2 and 3, 2.3 ng/g and 2.9 ng/g respectively, were detected.

The highest concentration of ΣDDT (102.3 ng/g) was found at the sampling station #1. This value is probably due to an explosion which occurred in 1982 in a store for pesticides, where the principal product kept was DDT; this store was located in the city Managua 1 km from the sampling station #1. This explosion may also have influenced station #2 where 23.0 ng/g ΣDDT were found. The large quantity of DDT which has been used in vector control programs in the city may also have influenced the levels at these two stations nearest the city.

In five lakes in the United States, Cooper et al[27] in 1979 found ΣDDT concentration from 226 - 1,276 ng/g in surface sediments. In another investigation, carried out in the North Sea, where one may suppose that no anthropogenic contamination exists, the ΣDDT average found was however 0.56 ng/g.[28] In one of the most polluted areas of the world (the Southern California Bight near Los Angeles), an average ΣDDT concentration in sediment of 94 μg/g was found.[29]



## CONCLUSIONS

The most prevailing pesticides in fish and sediments from Lake Xolotlán were toxaphene and ΣDDT. The Hercules factory still stores large quantities of toxaphene and the Nicaraguan agricultors keep on using this product. Due to this in the future it will be necessary to carry on investigating the concentration of toxaphene residues in our ecosystems. It would be useful to investigate the metabolism of DDT and toxaphene in Lake Xolotlán, to obtain a better knowledge of the transformations of these compounds in an aquatic ecosystem below unusual ecological conditions in tropical climate.

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