Bull. Environ. Contam. Toxicol. (1984) 33:423–429

Bulletin of Environmental Contamination and Toxicology

Relative Importance of Dietary and Environmental Sources of Lindane in Fish

C. Marcelle* and J.-P. Thomé

Laboratory of Animal Morphology, Systematics and Ecology, Zoology Institute, University of Liège; 22, Quai Van Beneden, 4020, Liège, Belgium

Recent surveys have shown the wide distribution of lindane in freshwater ecosystems in Belgium (Gordts 1975, Thomé and Thomé 1982, Marcelle and Thomé 1984). Although different studies have suggested that lindane is strongly accumulated in freshwater organisms (Portman 1979, Ullmann 1972), data about the relative importance of accumulation through the food or directly from the water are not available.

This study attempts to evaluate, under laboratory conditions, the relative importance of both modes of entry of pesticide into fish. These laboratory conditions were chosen to be representative of a natural ecosystem: the experiment was conducted for a long period of time, on a wild and widely distrbuted species, the gudgeon-*Gobio gobio (L.)*-, and in a water contaminated by lindane in the ppb range, i.e. concentrations found in belgian rivers running in agricultural area (Marcelle and Thomé 1984).

MATERIAL AND METHODS

In this experiment, the fish were fed with contaminated and non contaminated Chironomidae larvae. Fish and Chironomidae larvae were obtained from a local fishing store.

The gudgeons were 8 to 10 cm long and had an average weight of 6.1 g (σ = 2.1). Fish and larvae were distributed in 70 L tanks continuously supplied with 60 ml/min of lindane contaminated tap water. The lindane concentration was maintained at (1.1±0.2) ppb in well aerated water during the intoxication period.

120 fish were distributed in 4 tanks (30 fish per tank). Two tanks were designed for the intoxication of fish fed with Contaminated Food (CF tanks); the other 2 were designed for the intoxication of fish fed with Non Contaminated Food (NCF). Chironomidae larvae were maintained in a 5th aquarium. Uncontaminated larvae were placed in a small aquarium continuously supplied with tap water.

Fish were fed once every two days, during the intoxication period, with 6 to 3 g of larvae per tank, depending on the number of fish remaining in the aquariums. During the detoxication period, they were not fed.

*: present address: Rue du Mont, 24; B-4458 Fexhe-Slins, Belgium.

The physico-chemical parameters of water, measured during the experiment with an Aqua Merck kit were: pH = 8; total hardness: 200-220 ppm CaO; nitrites: 0.05-0.1 ppm; nitrates: 25-30 ppm; NH_4^+: not detected. Temperature was maintained at (14 ± 1)°C.

At different intervals, 3 fish were removed fron both the CF and the NCF tanks and lindane concentrations were determined; the lindane accumulation in larvae was also followed. Lindane concentrations were measured in water, brain, liver, and Chironomidae larvae as previously described (Marcelle and Thomé 1983).

Gudgeon and larvae lindane accumulation were followed for 18 days. The pesticide flow was then stopped and the lindane concentration was followed in the fish during a 30 days detoxication in tap water.

RESULTS AND DISCUSSION

The lindane concentration in Chironomidae larvae at the beginning of the experiment was 50 ppb; the concentration reached 100 ppb after a week of exposure. This concentration corresponds to what can be found in the usual food of gudgeons under natural conditions (Marcelle and Thomé 1984).

Table 1 gives lindane contents measured in liver and in brain at different times during a chronic intoxication with lindane and during the subsequent detoxication.

As in a previous work (Marcelle and Thomé 1983), the logarithmic transformation of the data was used before statistical analysis. We have shown that no significant correlation can be found between the total fresh weight of fish and the lindane content found in each analysed organ (liver and brain). This observation is opposed to the results obtained by different authors (Buhler et al. 1970, Kanazawa 1978). Our results could be explained by the weak dispersion of fish weights as a result of the fish selection made before the experiment.

As the sex of each fish was determined during dissection, statistical analyses were conducted in order to determine wether a correlation between lindane content and sex could be demonstrated (Figure 1). We found significant differences according to fish sex during both intoxication and detoxication.

a) during intoxication, the lindane accumulation in male fish liver was higher than in female fish liver ($p \leq 0.05$), while male fish brain accumulated less lindane than female fish brain ($p \leq 0.01$).
b) during detoxication, male fish liver showed a higher lindane content than female fish liver ($p \leq 0.03$); no significant difference in lindane contents of brain could be found between male and female fish.

In mammals, it has generally been shown that pesticide bioaccumulation was higher in males than in females (Matsumura 1975). We confirmed these observations on the gudgeon only in the case of liver, not for the brain.

Table 1: Lindane contents (in ppb) in gudgeon liver and brain during a chronic intoxication. Lindane concentration in water: 1ppb. Fish were fed with either Contaminated Food (CF) or Non Contaminated Food (NCF).

Sampling number	Time (h)	Liver[1]		Brain[1]	
0	0[2]	20 ± 4 (6)		91 ± 60	
1	6	85 ± 24 (5)		97 ± 45	
2	23	199 ± 86 (5)		70 ± 16	
	24[3]	NCF[4]	CF[4]	NCF[4]	CF[4]
3	30	184 ± 6	225 ± 93	65 ± 28	191 ± 103
4	49	330 ± 108	241 ± 4	219 ± 41	167 ± 38
5	74	420 ± 166	506 ± 234	207 ± 48	270 ± 135
6	126	431 ± 248	479 ± 355	116 ± 31	180 ± 61
7	220	1159 ± 609	341 ± 124	328 ± 38	1137 ± 791
8	315	1832 ± 1277	900 ± 240	461 ± 132	395 ± 79
9	433	1596 ± 1065	4037 ± 3883	759 ± 471	620 ± 228
----------	457	-------- End of intoxication	----------	----------	----------
10	536	205 ± 104	350 ± 330	92 ± 39	147 ± 51
11	728	136 ± 149	142 ± 38	65 ± 25	100 ± 25
12	1178	50 ± 6	65 ± 2	26 ± 11	54 ± 3

[1]: mean ± SD; (x): number of analyzed fish
[2]: lindane concentrations in the control group
[3]: time of the first feeding
[4]: for each moment, after the first feeding, results are the mean of analyses of 3 fish

During intoxication, a two-way analysis of variance (lindane concentration vs type of food) did not show significant differences in lindane accumulation in liver and brain of fish fed with either normal or intoxicated food. All the data were thus combined. We found for the brain a very highly significant linear correlation between the neperian logarithm of lindane concentration in this organ and the time of exposure to pesticide (Figure 2).

For liver, the correlation between both terms of relation was logarithmic.

These results implied that the decimal lindane concentration in brain increased exponentially with the time of exposure to lindane; on the other hand, lindane concentration in liver increased linearly with the time of exposure to the toxicant.

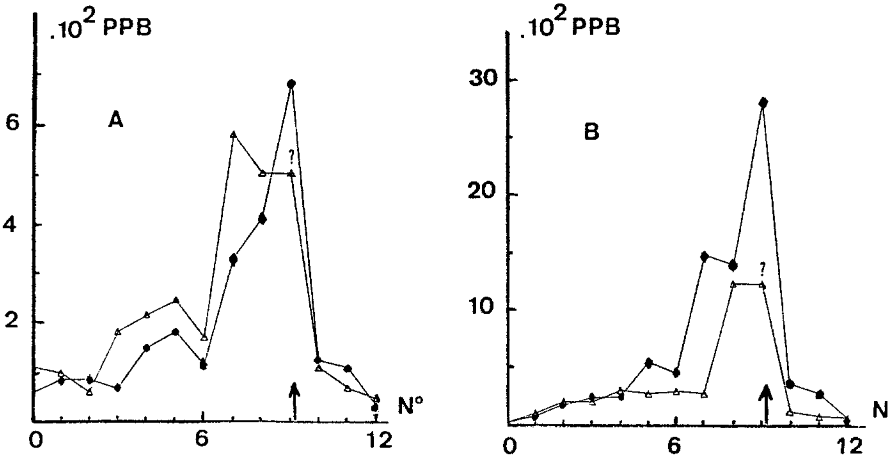

Figure 1: Means of lindane concentration in brain (A) and in liver (B) of male (♦) and female (Δ) fish during an intoxication and the subsequent detoxication. The beginning of detoxication is shown by an arrow.
N°: sampling n° (see table 1 for the times corresponding to sampling n°)
?: no female in this sampling; we show the result of the preceeding sampling.

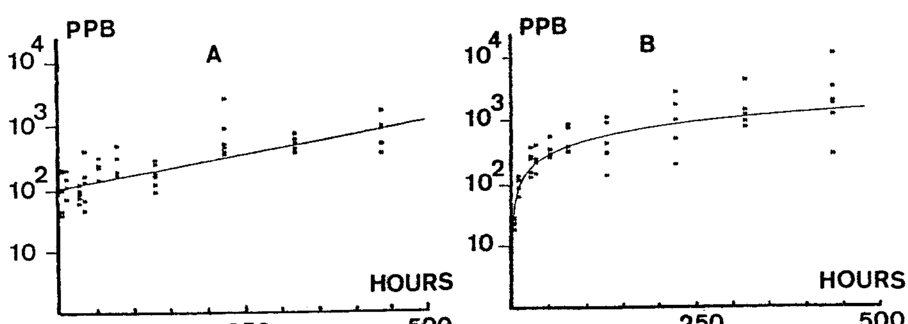

Figure 2: Evolution of lindane contents in brain (A) and in liver (B) during a chronic intoxication; CF results and NCF results are combined. Regression equations and regression coefficients are:
brain: $y = 0.005\ x + 4.52$; $r = 0.766$
liver: $y = 0.754\ \ln x + 2.619$; $r = 0.890$.

In a previous experiment (96 hours LC 50 experiment, Marcelle and Thomé 1983), we had shown similar increases of lindane contents in brain and liver with the lindane concentration in the medium.

Concentrations similar to those attained in the present experiment had been found in brain and liver after a 96 hours exposure to a lindane concentration in the medium of ± 37 ppb. Such a concentration in the medium had induced a 10% mortality in the fish population. With identical brain and liver contamination, we did not observe similar mortality in the present experiment. It seems that adaptation mechanisms, favorable to the fish survival, have taken place. One mechanism effective against pesticide penetration could be the blood-brain barrier (e.g. Morgan and Roan 1970, Davison 1970). This is supported by data from previous experiments (Marcelle and Thomé 1983) showing blood-brain barrier effectiveness at low (0.2-20 ppb) pesticide concentration in water.

For detoxication, a two-way analysis of variance of the results obtained for liver did not show significant differences in lindane contents found in liver of fish fed with normal or intoxicated food. Thus the detoxication rate in liver did not depend on the nature of food. Combining the data, we found a significant logarithmic correlation (p = 0.001) between pesticide content in liver and duration of detoxication (Figure 3).

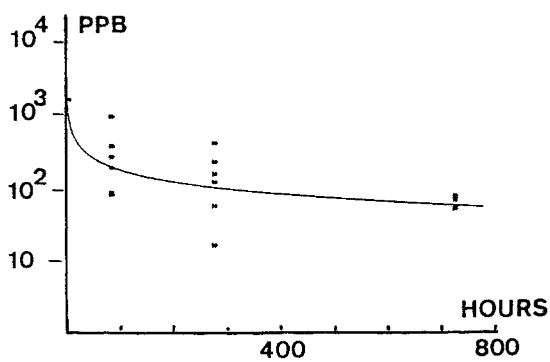

Figure 3: Detoxication curve of liver, after a chronic intoxication. CF and NCF are combined. Regression equation and regression coefficient are:
$y = -0.556 \ln x + 7.665$; $r = -0.706$.

However, in brain, detoxication rate seemed to depend on the type of food given during the intoxication period (Figure 4). When fish were fed with contaminated food, the detoxication in brain was slower than in fish fed with non contaminated food. In both figure 3 and 4, the concentration at the beginning of detoxication were calculated from the regression equation given in figure 2.

At the end of detoxication, the lindane contents in brain were lower than at the beginning of the experiment (35 ppb vs 60 ppb). In liver, after 31 days of detoxication, the pesticide contents were still higher than at the beginning of the experiment (60 ppb vs 20 ppb).

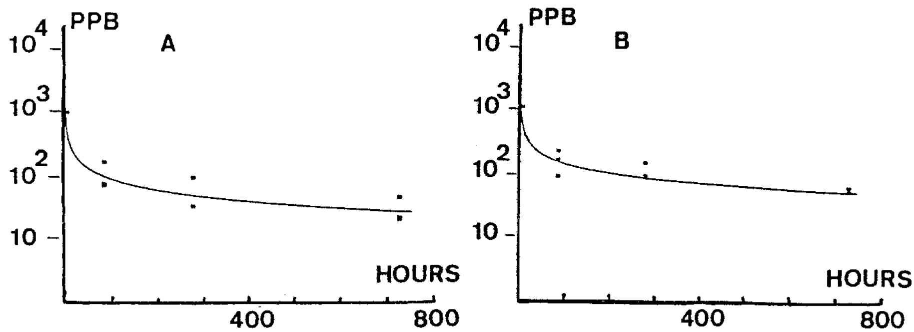

Figure 4: Detoxication curves of brain, after a chronic intoxication. A: fish fed with non contaminated food; B: fish fed with contaminated food. Regression equations and regression coefficient are:
A: $y = -0.582 \ln x + 7.125$; $r = -0.918$
B: $y = -0.482 \ln x + 7.167$; $r = -0.950$

It was obvious in our experiment of chronic intoxication that the transfer of pesticide via the food chain was of lesser importance to the accumulation of pesticide than the direct uptake from water. This could be due to the fact that the difference in lindane contents between contaminated and non contaminated larvae was too small (100 ppb vs 50 ppb). This conclusion is opposed to the results generally obtained for most lipophilic organochlorine insecticides such as DDT e.g. (Matsumura 1975, Edwards 1973). Our conclusion, however, is in accordance with those of Ellgehausen et al (1980), who have demonstrated, for less lipophilic pesticides such as atrazine, monuron, 2-4 D, the importance of the direct uptake from the environment.

Acknowledgements. Continuous support of Prof. Ch. Jeuniaux is gratefully acknowledged.

REFERENCES

Buhler DR, Shanks WE (1970). In: Edwards CA (1973)
Davison KL (1970). In: Matsumura F (1975)
Edwards CA (1973) Environmental Pollution by Pesticides. Plenum Press, London and New-York
Gordts A (1975) Inventaris van de Waterentreininging. Verlagen over de Voor Uitgang der Werken, 1974. 8. Waterpesticides. I.C.W.B. (I.H.E.) 9p, Brussel 1974
Kanazawa J (1978) Bioconcentration Ratio of Diazinon by Freshwater Fish and Snails. Bull Environ Contam Toxicol 20: 613-617.
Marcelle C, Thomé J-P (1983) Acute Toxicity and Bioaccumulation of Lindane in Gudgeon, *Gobio gobio (L.)*. Bull Environ Contam Toxicol 31: 453-458

Marcelle C, Thomé J-P (1984) Etude Analytique de la Contamination d'Invertébrés Aquatiques par le Lindane dans une Rivière de Hesbaye. Annales de la Société Royale Zoologique de Belgique (in press)
Matsumura F (1975) Toxicology of Insecticides. Plenum Press, London and New-York
Morgan A, Roan B (1970). In: Matsumura (1975)
Portman JE (1979) Evaluation of the Impact on The Aquatic Environment of HCH (HCH Isomers), HCB, DDT (+DDE and DDD), Heptachlor (+ Heptachlor Epoxide) and Chlordane. Commission of the European Communities, Env/486/79
Thomé J-P and Thomé M (1982) Enquête sur les Espèces de Vertébrés Menacés de Disparition en Wallonie, Tome VII: Les Pesticides et les Métaux Lourds comme Facteurs de Risques pour la Faune Sauvage, Ministère de la Région Wallonne pour l'Eau, l'Environment et la Vie Rurale
Ullmann E (1972) Lindane: Monograph of an Insecticide; Publ. Verlag K. Schillinger, Freibrug im Breisgau

Received January 26, 1984; accepted February 7, 1984