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The toxicity of pesticides to marine animals, and further data on the occurrence of pesticides in oysters

by

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Introduction

Although it is now widely acknowledged that pesticides can be toxic to marine animals and that the organo-chlorine group are only very slowly broken down, there is still very little knowledge about the levels which affect marine life, either in the water or in the animal itself. Most of the work relevant to the marine environment has been carried out in America. For example, at the Gulf Breeze laboratory pesticides are routinely screened for toxicity (Butler 1966), and each year large numbers of shellfish and fish are analysed for their pesticide content. Much of the information obtained is published annually by the U.S. Fish and Wildlife Service in a report on "The Effects of Pesticides on Fish and Wildlife" (see, e.g., Anon. 1964 and 1965). However, little of this work is directly applicable to our own part of the marine environment. In general the American species are different, and the ambient water temperatures usually differ from our own by at least 5°C and in winter often by as much as 20°C. Temperature and species differences can be important, as has been shown, for example, by Portmann (1968). For this reason the toxicity of some pesticides used in the United Kingdom is being examined at Burnham-on-Crouch. Some of the experimental results are reported in part 1 of this paper.

Since the pattern and degree of pesticide usage in the United States is very different from that in the United Kingdom, American pesticide residue results cannot in general be used as indicators of the levels likely to be found in marine species caught off the United Kingdom coast. During the past few years increasing attention has been paid to this problem and some results have now been published. Holden (1966) reported some results for marine fish, and more recently Holden and Marsden (1967) reported further data on seals, porpoises and fish. At last year's ICES meeting we reported some of the early

results of analyses made at Burnham-on-Crouch. Some further pesticide analysis results relating to oysters are reported in part 2 of this paper.

Part 1

Methods and Results

The method of toxicity testing employed in experiments with brown shrimps (Crangon crangon) and cockles (Cardium edule) is described briefly here (for further details see Portmann 1968).

All the experiments were carried out over forty-eight hours in a controlled temperature room at 15°C, using 10 litre perspex tanks. The room was in darkness throughout the experiment. Solutions of the pesticides were prepared by dissolving them in acetone, because they were not very soluble in water; diquat and paraquat were the only exceptions and these were diluted with distilled water. The concentrations of the pesticide solutions were such that the volumes added to the tanks did not exceed 10 ml. A range of concentrations of each pesticide in sea-water was set up, such that each concentration was three times as strong as the previous one, e.g. 0.001 ppm, 0.0033 ppm, 0.01 ppm, etc.

Fifteen tanks were used in the experiments and two of these were set up without any pesticide; these acted as controls. Acetone was added to all the tanks (except in experiments with paraquat and diquat) including the controls, so that each tank contained 10 ml acetone. The tanks were stirred, to ensure a homogeneous mix, the aerators turned on, and twenty-five specimens of the test species introduced to each tank. Dead animals were removed during the course of the experiments, and the total numbers which had died in each tank were recorded at the conclusion of each experiment.

The highest concentration at which less than 50 per cent of the animals died, and the lowest concentration at which more than 50 per cent died, were noted and the LC_{50} (concentration required to kill 50 per cent of the test animals) was said to lie between these two concentrations. In some instances the LC_{50} appeared to lie very close to a particular concentration and this accounts for the single figures given in the table of results below. The figures refer to the quantity of pesticide added to the sea water.

Pesticide		48 hr LC_{50} at 15°C (in ppm)	
Group	Under test	Crangon crangon	Cardium edule
Organochlorine insecticide	DDT	0.0033 - 0.01	>10
	BHC	0.0033	>10
	Dieldrin	0.01 - 0.033	>10
	Endosulfan	0.01	>10
	Tetradifon	>10	>10
Organo-phosphorus insecticide	Parathion	0.0033 - 0.01	3.3 - 10
	Malathion	0.33 - 0.1	3.3 - 10
	Azinphos-methyl	0.00033 - 0.001	>10
Bipyridyl herbicide	Paraquat	>10	>10
	Diquat	>10	>10

These results demonstrate that very low concentrations of organo-phosphorus and organo-chlorine insecticides are lethal to Crangon crangon. Cardium edule appears to be much less susceptible than Crangon crangon and is least resistant to organo-phosphorus insecticides; Crangon crangon also appears to be most susceptible to this class of compound.

In all the experiments except those with paraquat and diquat the possibility of the synergistic effect of acetone and the pesticide must be borne in mind. The fact that the acetone alone did not cause deaths in the controls does not necessarily mean that it does not impose a stress. Experiments are now planned which will investigate the influence of acetone. An additional unknown factor is the rate at which pesticide is lost from solution by precipitation, evaporation, breakdown or adsorption onto silt particles. In order to establish this, analysis of the waters would be necessary.

Duplicate experiments did not always give the same results and two main factors are believed to be involved here. The first is the variation in aeration. Aeration is carried out by means of diffuser stones controlled by

small nylon taps, and although each is carefully adjusted to give the same rate differences inevitably occur. It is believed that these could cause significant differences in results. The second factor involved is the variation in size and condition of the test animals. The size is now standardized but condition is much more difficult to control, because it varies with season etc. Where duplicate experiments did not agree the experiments were repeated until a consistent result was obtained.

Conclusion

Very low concentrations of organo-phosphorus and organo-chlorine pesticides can be lethal to some shellfish. Organo-phosphorus compounds appear to be slightly more toxic than organo-chlorine compounds. The results of the toxicity experiments reported above are subject to a number of variables which could affect results. Steps are now being taken to eliminate as many of these as possible.

Part 2

Methods and Results

Oysters have been analysed for their organo-chlorine pesticide content since March 1967. These analyses have been carried out on batches of 5 oysters (Ostrea edulis) collected at two-weekly intervals from two local oyster grounds, one in the Roach estuary and one in the Crouch estuary.

The tissue from each sample of 5 oysters was bulked and homogenized, and two 8g sub-samples were then mixed with anhydrous sodium sulphate. Duplicate 100 ml extracts were then prepared by repeatedly simmering the mixes with hexane. Clean-up of the hexane extracts was by the method of de Faubert Maunder et al. (1964), and subsequent analysis was by gas-liquid chromatography. The gas chromatograph used was a dual column dual electron-capture detector instrument. The columns used were 5 ft $\frac{1}{8}$ inch O.D. glass and were packed with 2.5 per cent SE301 + 0.25 per cent. Epikote 1001 on Chromosorb G and 2 per cent Oronite polybutene 128 + 0.2 per cent Epikote 1001 on Gas Chrom Q.

Each sample was analysed for α, β and γ BHC, heptachlor, epoxide, aldrin, dieldrin, endrin, DDT, TDE and DDE. Of these compounds α and γ BHC, dieldrin, DDT, TDE and DDE were found in most samples. The results of the two analyses for each sample of oysters were very often the same and were usually within 10 per cent of each other, which is as good as the methods used will permit. Where the results of the two analyses differed the mean of the two values was taken.

The results of the two-weekly analyses for the Crouch and Roach oysters collected during 1967 are shown in Figures 1 and 2 respectively. In order to give a clear picture the concentrations of the two BHC isomers were combined and plotted, as were the concentrations of dieldrin, DDT and its two metabolites, TDE and DDE. A third line shows the concentration of dieldrin alone and the fourth line shows the total concentrations of all pesticides.

Butler (1966) showed that oysters are good indicators of organo-chlorine pesticide pollution, since they can rapidly concentrate these materials and, after the pollution is past, excrete them. The two sets of figures for the Crouch and Roach oysters followed a similar pattern for most of 1967, demonstrating a similar pattern of pesticide pollution. The only major difference occurred in June-July, when the rise in pesticide concentration was much greater in the Roach oysters than in those from the Crouch. A second peak in October-November was shown by both sets of oysters. Throughout the remainder of the year the levels were comparable and fairly steady.

The peak in October-November is in both instances probably due to pesticide usage on autumn-sown wheat. BHC and dieldrin are widely used as seed dressings, on wheat sown during the autumn, as a protection against attack by wheat bulb fly and certain seed-borne diseases. The parallel increase in DDT group concentrations is not so readily accounted for, and as yet no explanation has been found.

The large peak in all pesticide concentrations found in late June in Roach oysters could be the result of pesticide usage in horticulture. Dieldrin, DDT and BHC are all used against a variety of pests in orchards, on soft fruits and in market gardens at this time of year. In the Roach catchment area there is a large acreage of market gardens and a significant area planted with black-currants, raspberries and strawberries. In the Crouch catchment area there is only a small area of market gardens and strawberry fields, and most of the land is under wheat or barley which would not receive pesticide treatment at this time of year. This difference in horticultural and agricultural practice could explain the difference in pesticide concentrations found between Roach and Crouch oysters.

It is now generally accepted that organo-chlorine pesticides are largely adsorbed by soil particles and are not washed off the soil, by rainfall, into the rivers (Strickland, personal communication). This poses a problem as to how the pesticides do get into the oysters. There are in fact three possible ways in which this could happen:

a) The pesticides would slowly evaporate and could be rained out into the rivers. Since the peaks occur very shortly after the suspected period of usage this can only be a minor contributory factor.

b) The pesticides may be washed into the rivers in the course of the washing out of containers or equipment used in applying them. This could be the main contributory factor.

c) It is possible that soil particles carrying adsorbed pesticides are washed into the rivers and that the oysters filter some of the pesticide-contaminated solids during feeding, desorbing the pesticide in the process. It is not known how important this might be.

Conclusion

It appears that oysters concentrate and release pesticides according to the quantity of pesticide in the water. A fairly constant background level is maintained throughout the year but peak concentrations occur in early summer and autumn. These peaks are probably the result of seasonal agricultural or horticultural usage. Further analyses have been carried out during the present year and the programme will continue during 1969. It is intended to report on the results of the 1968 and 1969 analyses at a later date.

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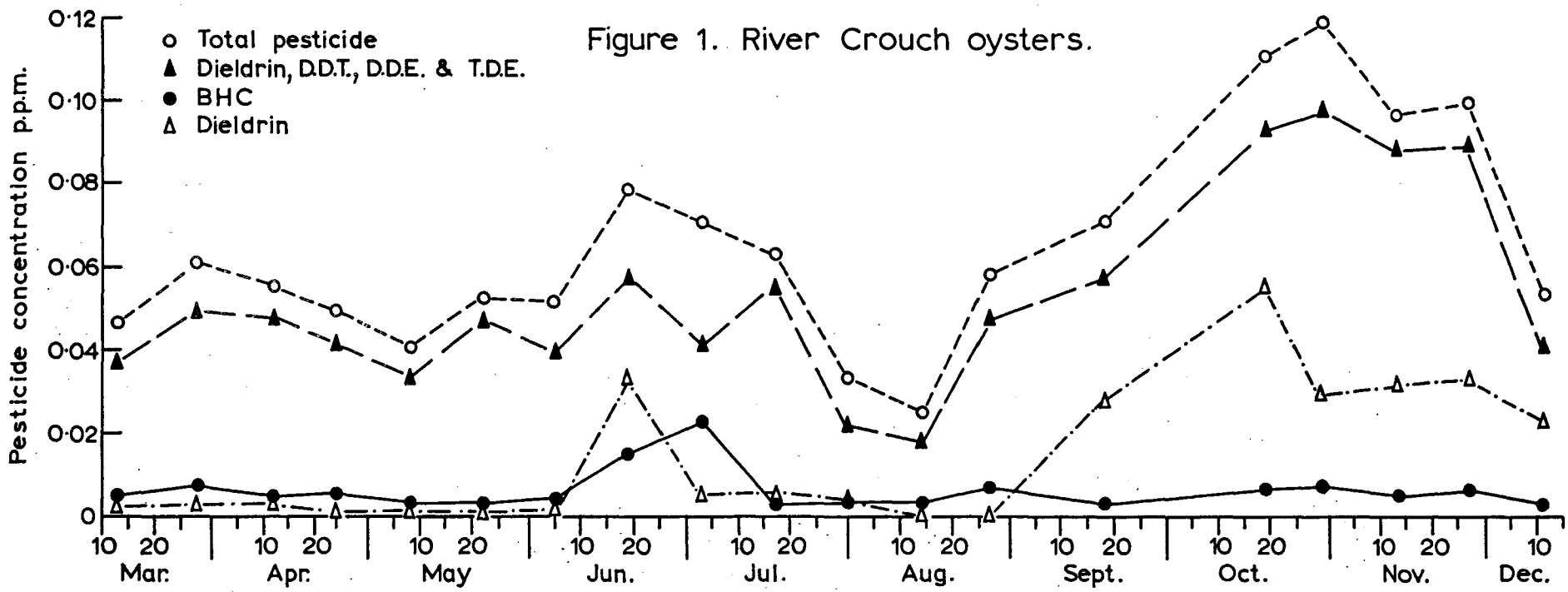

Figure 1. River Crouch oysters.

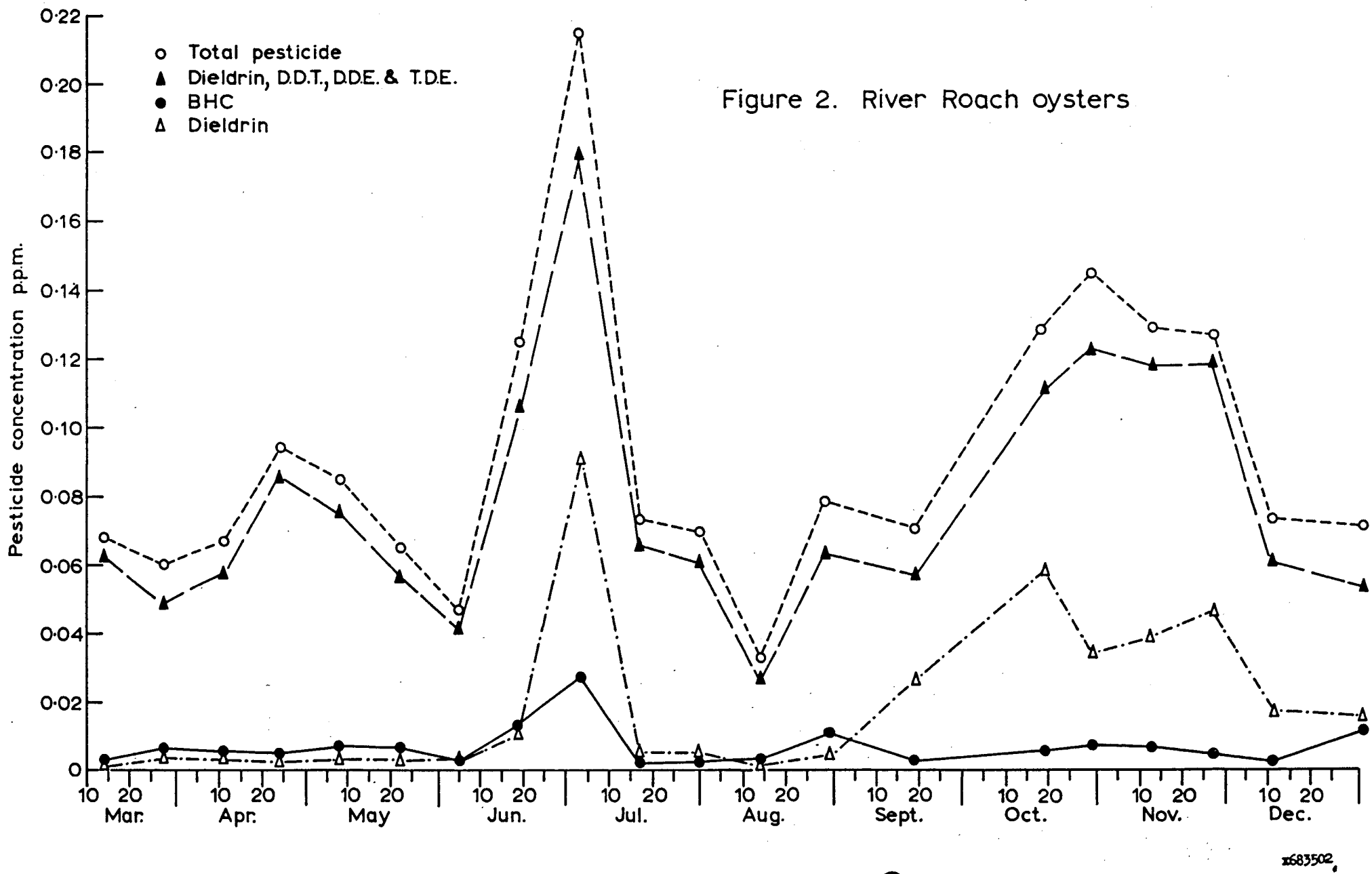

Figure 2. River Roach oysters