Temperature Selection in Brook Trout (*Salvelinus fontinalis*) Following Exposure to DDT, PCB or Phenol[1]

by D. L. Miller and D. M. Ogilvie

Department of Zoology
University of Western Ontario
London, Canada N6A 3K7

Introduction

Synthetic chlorinated hydrocarbon compounds such as dichlorodiphenyltrichloroethane (DDT) and polychlorinated biphenyls (PCB's) are now widely distributed throughout the biosphere. Because of similarities in their molecular structure, it is somewhat difficult to chemically distinguish DDT from a PCB and indeed, the presence of the latter may interfere with pesticide residue analysis (REYNOLDS 1969). The ubiquitous nature of these pollutants has raised many questions relating to their possible physiological and ecological effects (WURSTER 1969; GUSTAFSON 1970).

In the case of DDT, it has been shown that this chemical can interfere with behavioural thermoregulation (temperature selection) in fish (OGILVIE and ANDERSON 1965; GARDINER 1973), and the ecological implications of this finding have been considered by several authors (WURSTER 1969; WARREN 1971). An investigation of temperature selection in salmonids following exposure to a variety of chlorinated hydrocarbons revealed that although methoxychlor and several DDT analogs all shifted the selected temperature in a similar manner, the response was not altered by exposure to a PCB (PETERSON 1973).

In view of the chemical similarity between DDT and the PCB's, the apparent insensitivity of the temperature selection mechanism to PCB is of particular interest. Since Peterson reported on the effects of only one PCB dosage, however, the finding is not yet well documented. This paper therefore describes a more extensive comparison of the effects of DDT and a PCB on temperature selection in the brook trout. In addition, we also report on the effects of phenol. This chemical is of interest because as a pollutant it is dangerous to fish (JONES 1964), and in addition, its simple chemical structure is the basic component of both DDT and the PCB's.

1. Based on a thesis submitted by the senior author in partial fulfillment of the requirements for the degree of Master of Science at the University of Western Ontario.

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Bulletin of Environmental Contamination & Toxicology, Vol. 14, No. 5 © 1975 by Springer-Verlag New York Inc.

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Methods

Underyearling brook trout (Salvelinus fontinalis, Mitchell) two to three months old were obtained from a commercial trout hatchery and held at 15°C for at least two weeks prior to experimental use. The fish were subjected to a 12L:12D photoperiod, and were fed Ralston Purina trout food once a day. Stock solutions of technical grade DDT (Nutritional Biochemical Corporation) were prepared in an acetone carrier, while the polychlorinated biphenyl (Monsanto Company, Arochlor 1254^{R}) was dissolved in dimethylsulfoxide. Phenol (BDH Chemicals) was dissolved in water, and because of its volatile nature fresh solutions were prepared daily.

Prior to temperature gradient tests, the trout were fed for 10 minutes and then exposed for 24 hours to DDT (10 to 50 ppb), phenol (0.75 to 10 ppm), or PCB (25 to 100 ppm). For each exposure groups of five fish were placed in 2.5 litres of gently aerated water maintained at 15°C and containing 2.5 ml of the test chemical plus its carrier, or 2.5 ml of the carrier only. For each concentration tested two groups of experimental fish and two groups of control fish were so exposed and then tested simultaneously in a multichannel thermogradient device.

The temperature gradient apparatus has been described previously (OGILVIE and FRYER 1971). For these experiments four lanes were used, each 13 cm wide, 190 cm long, and 4 cm high. Each lane was divided into 20 positions 9.5 cm wide. Immediately after their exposure, each group of fish was placed in a separate lane and temperature selection in the control and experimental fish was determined simultaneously. Because some mortality occurred during exposure to the higher doses of the test chemicals, the number of experimental fish varied from 11 to 20.

Water depth in the apparatus was maintained at 1.8 cm and temperatures were recorded 5 mm above the floor of the control and experimental lanes with a YSI model 44TD telethermometer and a type 402 thermistor probe. Temperatures were found to be relatively linear along the length of the apparatus and ranged from about 4°C to 29°C. Gradient temperatures were recorded immediately before the introduction of the fish into the apparatus and at the completion of an observation period.

During a temperature selection experiment, the fish were viewed indirectly by means of a mirror

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suspended over the apparatus with the observer located behind a cardboard blind. After a one-hour equilibration period, the locations of the fish were recorded every two minutes for one hour.

The data was analyzed by arbitrarily dividing the thermogradient into 2°C intervals and determining the number of observations in each interval (see Fig. 4). Mean selected temperatures were then calculated and compared by Student's t-test. Differences were considered statistically significant when the P value was less than 0.05.

Results and Discussion

In the DDT experiments (Fig. 1) the mean temperatures selected by fish which had been exposed to the acetone vehicle only were relatively constant, ranging from 12.9 to 13.3°C. In the case of the DDT-exposed fish, however, the lowest dose used (10 ppb) significantly depressed the mean selected temperature, while the highest dose (50 ppb) significantly increased the selected temperature by more than four degrees. These results are clearly indicative of the biphasic dosage-response relationship reported previously for brook

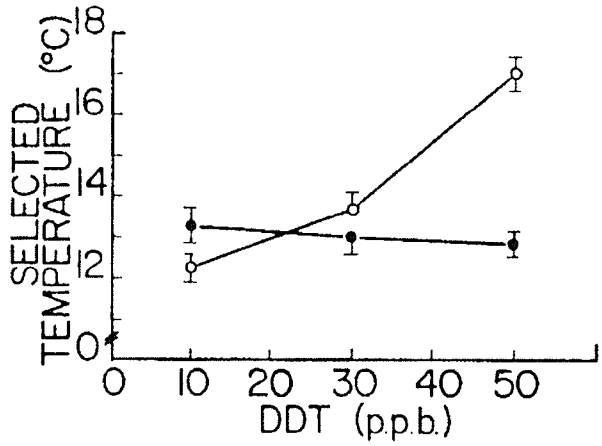

Fig. 1. Mean temperatures selected by brook trout following a 24-hour exposure to DDT in an acetone carrier (open circles) or acetone only (closed circles). Vertical bars denote ± 2 S.E.M.

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trout (GARDINER 1973) and also for Atlantic salmon (OGILVIE and ANDERSON 1965).

On the basis of the mortality which occurred during the DDT exposures, the 24-hour LD_{50} was estimated to be 54 ppb. No mortality occurred at 10 ppb, however, exposure to 30 ppb caused 20% mortality, and 50 ppb killed 45% of the fish in 24 hours. Thus, although the maximum shift in selected temperature was associated with relatively high mortality, the significant decrease in selected temperature produced by exposure to 10 ppb was not associated with mortality and therefore represents a truly sublethal effect.

It is evident from the phenol experiments (Fig. 2) that the control fish for the 10 ppm exposure selected a significantly higher temperature than the 0.75 and 5.0 ppm controls. The reason for this is unknown, however, we have observed similar day-to-day variation in control selected temperatures in other experiments (MILLER 1974), and the value of a multichannel thermogradient device in which control and experimental fish can be tested simultaneously is therefore apparent. Despite an unexplicable increase in the mean temperature selected by control fish, it is evident in Fig. 2 that exposure to phenol produced a consistant downward shift in selected temperature and further, this decrease was significant at doses of 7.5 and 10 ppm.

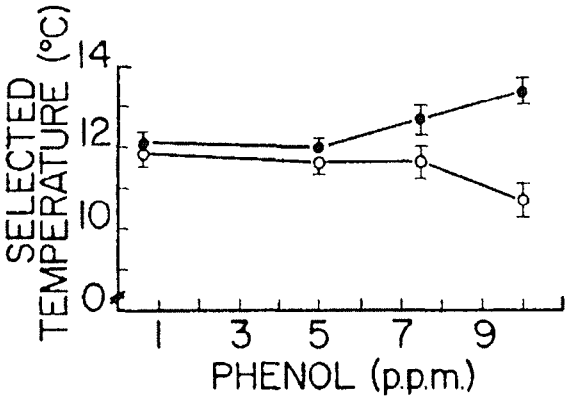

Fig. 2. Mean temperatures selected by brook trout following a 24-hour exposure to phenol (open circles). Control fish (closed circles) were held under comparable conditions for 24 hours prior to being simultaneously tested in the temperature gradient. Vertical bars denote ± 2 S.E.M.

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The 24-hour LD_{50} for phenol was estimated at 11.7 ppm and although the 10 ppm exposure produced 30% mortality, no deaths resulted from exposure to 7.5 ppm. As was the case with exposure to 10 ppb DDT, the temperature selection shift associated with this dose therefore clearly represents a sublethal effect.

As far as we are aware, there have been no previously published reports describing the effects of phenol on temperature selection in fish. It may be worthwhile to note, however, that we have obtained similar results with rainbow trout (MILLER 1974).

In contrast to the DDT and phenol results, 24-hour exposures to doses of PCB ranging from 25 to 100 ppm produced no mortality. The chemical similarity of DDT and PCB is well known (REYNOLDS 1969), and it is therefore somewhat surprising that no mortality was produced by PCB doses some thousandfold greater than the maximum DDT exposure used in these experiments. However, a separate mortality study using several species confirmed the fact that PCB is much less toxic to salmonids than DDT and in addition, the onset of mortality is somewhat delayed (MILLER 1974).

It is evident from Fig. 3 that all doses of PCB tested had no effect on the selected temperature of brook trout. Since these same exposures also produced no mortality, it may be argued that the fish were not subjected to a level of the chemical high enough to affect the mechanism of temperature selection. Since the maximum shifts in selected temperature produced by both DDT and phenol were associated with doses that also caused mortality, it seemed reasonable to increase the PCB exposure time until some mortality occurred, and then test the surviving fish in the temperature gradient.

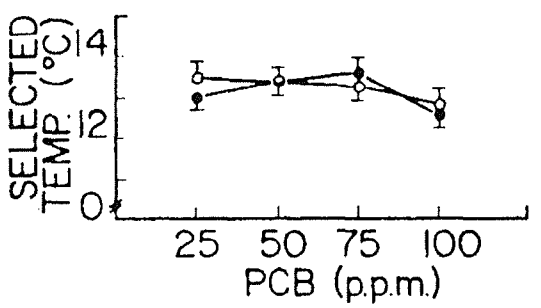

Fig. 3. Mean temperatures selected by brook trout following a 24-hour exposure to PCB in a dimethylsulfoxide carrier (open circles), or to dimethylsulfoxide only (closed circles). Vertical bars denote ± 2 S.E.M.

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Consequently, when 18 fish were exposed to 100 ppm. PCB for 48 hours, two fish died. The survivors were then tested in the gradient along with 19 controls. As can be seen in Fig. 4, a PCB exposure sufficient to cause mortality still had no effect on the selected temperature. In addition to the mortality noted above, a marked depression in locomotory behaviour was also observed during the 48-hour PCB exposure. Thus, while control fish swam actively about the exposure beaker, the PCB-exposed fish rested quietly on the bottom. Despite the chemical similarity between DDT and PCB, it therefore appears that PCB is much less toxic to fish than DDT and in addition, it does not appear to possess the ability to alter the thermoregulatory behaviour of fish as does DDT. This finding therefore confirms the preliminary observation by PETERSON (1973) who also failed to note any alteration in the temperature selected by PCB-exposed Atlantic salmon.

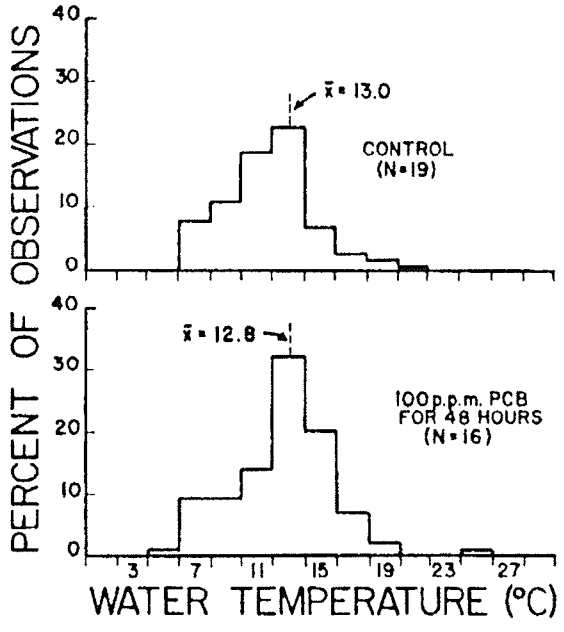

Fig. 4. Distribution of brook trout in a horizontal temperature gradient following a 48-hour exposure to 100 ppm PCB.

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