

## Further Studies on the Toxicity of DDT to Planaria

H. H. Kouyoumjian\* and J. P. Villeneuve

National Council for Scientific Research, Marine Research Centre,  
P. O. Box 11-8281, Beirut, Lebanon

Some aspects of the toxicity of p,p'-DDT and two of its metabolites p,p'-DDD and p,p'-DDE to the freshwater planarians Polycelis felina and Crenobia alpina were discussed by KOUYOUMJIAN et al. (1974a,b). These organochlorine pesticides were shown to inhibit rate of asexual fission and affect the coordinated movements of these animals. Planaria were shown to respond consistently to experimental treatment and that there seemed to be considerable interspecific variation in resistance to organochlorine chemicals and that intraspecific variation amongst individuals of the same size was low. Of the three organochlorine chemicals tested, DDD was the most toxic chemical to both species. Subsequently, it was also shown that these animals and a third species Phacogata velata could metabolize DDT to DDD and DDE (PHILLIPS et al. 1974, KOUYOUMJIAN et al. 1974b).

The present paper studies the possible deleterious effects of DDT on regeneration and asexual fission, and looks briefly into the rate of absorption and metabolism of DDT by normal and regenerating (decapitated) planaria.

### MATERIALS AND METHODS

Colonies of P. felina and C. alpina were kept in darkness, in aged tap water at  $12.0 \pm 0.5^{\circ}\text{C}$  (water temperature in the stream at the time of collection was about  $8.5^{\circ}\text{C}$ ). The water was changed regularly and the animals were fed weekly with washed beef liver to satiation. Experimental animals were starved 7 days preceding experimentation. Test animals were placed in p,p'-DDT (Ralph N. Emanuel Ltd.) with 1% acetone as carrier solvent. The number of survivors and fragments resulting from fission were recorded regularly.

The animals were decapitated by placing them on a wet slide and cutting off the heads just after the auricles with a sharp blade. Test animals were removed from the experimental dishes at 0.25, 0.5, 1, 2, 3, 4, 7, and 14 days, and analyzed for their contents of organochlorine chemicals. At the end of the experimental period, the animals were transferred to pesticide free, aged tap water for 7 days and then analyzed.

The analysis was accomplished by extraction with aceto-

nitrile, partitioning in hexane and finally cleaning on a florisil column. GLC analysis was done on a Varian Aerograph model 2100, on a 0.6 m, 2mm i.d. glass column packed with 10% OV-101 on Chromosorb W. The operating parameters were: Oven T. 200°C, Detector T. 250°C, Inlet port T. 205°C. The carrier gas was nitrogen kept at a constant flow rate of about 40 ml/min. The recovery of organochlorine pesticides using this technique was about 90-95%.

## RESULTS AND DISCUSSION

Initial GLC analyses of planaria revealed relatively low levels of organochlorine pesticide residues - 0 ug/g for DDT, 0.02 ug/g and 0.04 ug/g for DDD and DDE, respectively. Planaria generally responded well to experimental treatment and no deaths were reported throughout the experimental period in both control and experimental groups. The carrier solvent acetone was previously shown not to have any permanent deleterious effects at a concentration of 1% (KOUYOUJIAN et al. 1974a).

Figure 1 illustrates the inhibitory effect of DDT (0.15 ppm) on asexual fission in P. felina under the stimulatory effect of elevated experimental conditions - 12°C versus 8.5°C in nature at the time of collection.

Table 1 shows the rates of absorption and metabolism of p,p'-DDT by P. felina. It appears that during the first 24h of exposure to 0.15 ppm test chemical, decapitated planaria are able to resist DDT absorption with subsequent low levels of DDD and DDE. The lag in DDT absorption by decapitated planaria during the first 24h could be associated with concomitant decrease in permeability to the test chemical brought about by increased metabolic rates. HYMAN (1951) has reported that cutting planaria into small pieces accelerates the rate of oxygen consumption - a 24h stimulatory effect. After 24h, however, regenerating planaria absorb DDT at an increased rate presumably due also to the experimental body injury. DDT absorption by regenerating planaria appears to slow down the rate of regeneration, as shown by the fact that at the end of a 2-week period 4 out of a group of 10 planaria (C. alpina) kept in 0.4 ppm DDT (2 orders of magnitude below 96h LC 50) test solution had regenerated lost heads with 2 eye spots compared with 8 out of a group of 10 controls kept in 1% acetone solution.

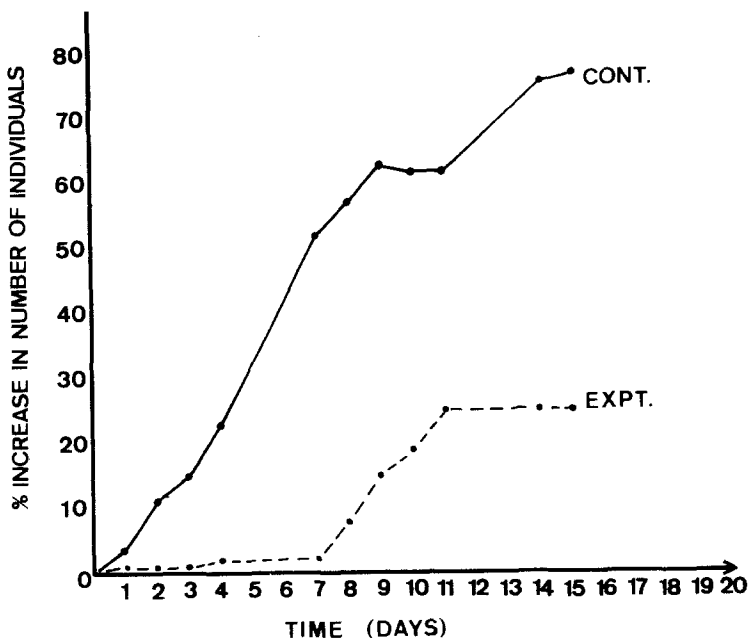


Fig. 1. Rate of asexual fission of P. felina in 0.15 ppm DDT (EXPT.) and 1% acetone (CONT.). 100 animals in each group.

It is shown here (Table 1) that normal and regenerating planaria can metabolize DDT to DDE and DDD and that DDE (dehydrochlorination) seems to be the major metabolite. PHILLIPS et al. (1974), working with P. velata have shown DDD (reductive dechlorination) as being the major product. In view of the higher toxicity of DDD to planaria, the latter situation might put any population containing high levels of DDD under considerable stress. It is also indicated that when exposed to sublethal concentrations of DDT, planaria could slowly absorb higher levels of DDT from the water with subsequent high body burdens of DDE and DDD than the respective 96h LC 50s of these test chemicals.

TABLE 1

DDT Absorption and Metabolism by Normal and Regenerating  
*Planaria* (*P. felina*)

| TIME  | DDT* |      | DDD  |      | DDE  |      |
|-------|------|------|------|------|------|------|
|       | CONT | EXPT | CONT | EXPT | CONT | EXPT |
| 0 h   | 0.00 | -    | 0.02 | -    | 0.04 | -    |
| 6 h   | 1.59 | 0.65 | 0.05 | 0.05 | 0.09 | 0.05 |
| 12 h  | 4.70 | 0.47 | 0.53 | 0.03 | 0.93 | 0.04 |
| 1 day | 8.56 | 0.62 | 0.86 | 0.24 | 1.80 | -    |
| 2 "   | 10.5 | 15.8 | 0.56 | 0.41 | 1.26 | 0.91 |
| 3 "   | 23.8 | 17.5 | 0.90 | 1.10 | 0.79 | 3.23 |
| 4 "   | 36.9 | 22.8 | 0.72 | 1.16 | 1.34 | 4.47 |
| 7 "   | 80.6 | 79.0 | 1.36 | 1.03 | 1.79 | 1.32 |
| 14 "  | 95.3 | 77.5 | 3.14 | 2.77 | 9.18 | 4.47 |

\*All concentrations in ug/g wet weight. Each value represents the mean of 3 determinations (3 groups of 10 animals each).

In view of the inhibitory effects of DDT on asexual fission and apparently on eye regeneration, it would be interesting to study the deleterious effects of such hazardous chemicals on brackish or marine flatworms and other organisms capable of regeneration - a process hitherto not looked into in great detail in this respect.

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