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The toxicity of DDT to the eastern brook trout, *Salvelinus f. fontinalis* (Mitchill)[1]

by

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Publicity describing the "miracles" of DDT has been universal following its recognition, during the war years, as an unusually effective insecticide. Its widespread popularity has led many biologists to utter words of caution against its indiscriminate use, since experimentation has shown this substance to be toxic to both warm- and cold-blooded animals. During the past 3 years, as a result of intensive studies, many articles have been published pertaining to the toxicity of DDT to various organisms. Roark (1944) submitted a review of the literature covering articles pertaining to the chemical and toxic nature of the substance from 1873 to 1944, and later (1944, 1945a, 1945b) assembled a list of publications and their respective authors.

The tests presented in this paper were started in early 1944, using the eastern brook trout *Salvelinus f. fontinalis* (Mitchill) as the test animal, at a time when little information as to the toxicity of DDT to fishes was available. Anonymous reports indicated that it was highly toxic to fishes; and since in mosquito control work and possibly in the control of other aquatic and terrestrial insects

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[1] Contribution from the Institute for Fisheries Research of the Michigan Department of Conservation.

conditions certainly would arise whereby fishes would be subjected to DDT, it was deemed desirable to examine this substance in its relationship to fishes.

In order to determine the degree of toxicity of DDT to our test animals and the amount of variation in toxicity, if any, resulting from various commercial methods of compounding, it was necessary to use several branded DDT compounds, prepared for a variety of specific uses. A total of 5 commercial products, (Table 1) supplied by Geigy Inc., New York, and one preparation mixed in our own laboratory, were used in these tests. The first three compounds listed in Table 1, containing petroleum and plant oil distillates, were designed specifically for the control of plant pests; the last three were compounded for the control of insects directly affecting man and animals. Their available DDT content ranged from 5 to 40 percent, and their reaction with water varied from a suspension to a tight emulsion.

Experimental Methods

The trout used in the tests, obtained from the Federal Fish Hatchery at Northville, Michigan, consisted of 7- to 9-months-old fingerling brook trout ranging from 2.5 to 5 inches in length. Since holding facilities were not available in the laboratory, a major portion of the stock supply of fish was retained at the hatchery in one of the nursery troughs. The water used in preparing and diluting the stock solutions was obtained from Flemming Creek, Washtenaw County, Michigan, a stream containing fish population and water characteristics comparable to those of the average spring-fed southern Michigan stream. The pH varied from 7.8 to 8.2 and the total hardness (i.e., methyl orange alkalinity) ranged from 188 to 200 p.p.m.

Table 1.--DDT products used in toxicity tests with the brook trout as the test animal. The first five products were supplied by Geigy Inc., New York, the last was prepared in our laboratory.

Name of product	DDT content (percentage)	Nature of base	Reaction with water	Specific use
Gesarol Oil Spray SHN20	20	Petroleum distillate	No emulsion	Control of plant pests
Gesarol Oil Spray SE20	20	Petroleum distillate	Loose emulsion	Control of plant pests
Gesapon 18	5	Plant oils	Tight emulsion	Control of soil insects
Neocid Larvacide	35	Petroleum distillate	No emulsion	Control of insects affecting man and animals
Neocid Barn Spray	10	Inert powder	Insoluble	Control of insects affecting man and animals
DDT plus kerosene	5	Petroleum distillate	No emulsion	Control of mosquitoes and domestic flies

The experimental test equipment consisted of one-gallon glass jars, covers, individual air dispersers, and a temperature control unit (Figure 1). To maintain a temperature of 61° to 65° F. in the test media during the runs, the jars were three-quarters immersed in a trough containing cold water which was in turn provided by circulating city water through brine coils retained within a deep-freeze unit. By regulating the incoming flow, the desired temperature was maintained.

Each test medium was made up to 3,000 cubic centimeters and aerated 20 minutes before the fish were introduced, to assure an adequate supply of dissolved oxygen at the start and to provide a reasonable degree of time for uniform dispersal of the test product. Following the introduction of the fish (5 in each jar) aeration was continued for a period of 96 hours (duration of test) or at least as long as any fish remained alive in the test medium. In cases where results were unexpected or inconsistent, tests were repeated to verify or refute the findings. The measurement of the degree of toxicity of the specific substances to fishes was attained by initial tests with widely differing concentrations, followed by more precise tests falling within the range of concentrations which killed or did not kill the fish in a 96-hour test period. The values derived were expressed in terms of consistent tolerance limits which represent the highest concentration of the test material at which all of the fish survived a 96-hour exposure.

Results

The toxic effect of DDT to brook trout, irrespective of the type of base employed in compounding, was similar in all respects. Several hours of exposure of the test animals in moderate DDT concentrations were required to produce the slightest symptoms. The first discernible

Figure 1.--View of laboratory equipment used in DDT toxicity tests, showing temperature control unit and arrangement of test jars.

reaction of the fish was a slight twitching of the paired fins and a jumpy movement of the opercles. The spasms lasted only a few seconds and were evenly spaced at 10- to 15-minute intervals. Upon continued exposure, the twitchy reactions became more pronounced, of longer duration and with increasing frequency. The final stage preceding death can best be described as a series of convulsions, followed by complete exhaustion of the test animal at which time the fish had lost equilibrium and the respiration was very weak and uneven. This period of quiescence (the precursor of death) was found to vary from 1 to 8 hours, depending upon the concentration of DDT employed in the test solution.

The DDT products designed specifically for the control of plant pests, namely: Gesarol SHN20, Gesarol SH20 and Gesapon 18, were found to possess different toxic values against fish. Gesarol SH20 was determined to exert twice the toxicity of Gesarol SHN20 and about six times the toxicity of Gesapon 18 (Table 2). Although the two Gesarols differed consistently in their tolerance limits, they showed a marked similiarity in respect to the velocity of toxicity to brook trout as compared to that of Gesapon 18. At concentrations of 0.40 p.p.m. DDT, Gesarol SHN20; and 0.20 p.p.m. DDT, Gesarol SH20, some of the trout died within 16 to 22 hours, while at concentrations of 0.20 and 0.10 p.p.m. DDT respectively, all of the fish survived the 96-hour test. In contrast, the minimum time required to produce death of the fish in various concentrations of Gesapon 18 ranged from 24 hours in a 2.0 p.p.m. DDT concentration to 67 hours at a concentration of 0.8 p.p.m. In other words, no sharp break, as displayed by the two Gesarols, occurred between lethal and sub-lethal concentrations of Gesapon 18.

Table 2.--Hours survival of brook trout in different concentrations of oil-base DDT products compounded to form loose and tight emulsions with water and specifically designed for use in plant pest control.

Concentration of DDT (p.p.m.)	Number of test animals used for each product	Products tested								
		Gesarol SM20 [1]			Gesarol SM20 [2]			Gesapon [3]		
		Hours survival			Hours survival			Hours survival		
		Minimum	Maximum	Average	Minimum	Maximum	Average	Minimum	Maximum	Average
4.0	10	16	40	33	18	33	28	2	15	14
2.0	10	16	63	38	17	34	26	24	74	48
1.0	10	16	90	32	18	33	23	39	96	88
0.8	10	26	96	67.2	17	33	23	67	96	91
0.6	10	16	96	42.4	18	32	26	96	96	96
0.4	10	16	96	58	18	33	26	96	96	96
0.2	10	96	96	96	22	68	44	96	96	96
0.1	10	96	96	96	96	96	96	96	96	96
0.08	10	96	96	96	96	96	96	96	96	96
0.04	10	96	96	96	96	96	96	96	96	96
0.00	10	96	96	96	96	96	96	96	96	96

[1] Petroleum distillate base containing no emulsifying agent

[2] Petroleum distillate base forming a loose emulsion

[3] Plant oil base forming a tight emulsion

Since it has been demonstrated that a difference in toxicity exists between the three products and that this difference was not accounted for in terms of the respective basic DDT content (Table 2), it appears that some of the toxicity displayed must have been due to the types of solvents and carriers employed in the compounding. Inasmuch as Gesapon 18 was the least toxic of the three products and was the only one which contained a plant oil base (the other two carrying petroleum distillate bases, Table 1) it is possible to conjecture that the plant oil was less toxic to the test animals than were the petroleum products. Another consideration which may also have exerted some influence in the variation of toxicity is the relationship between the products and their miscibility with water. Gesapon 18 is compounded in such a manner as to produce a tight emulsion in water; Gesarol SH20 forms a loose emulsion, and Gesarol SHM20 is immiscible in water. Surber (1946) found that DDT products producing an emulsion with water were more toxic to fishes than those in an oil solution containing no emulsifying agent. At a glance it does not appear that the results of these studies are in agreement with Surber's findings; Gesarol SHM20 (non-emulsifiable) was about three times more toxic than Gesapon 18 (tight emulsion) and only about one-half as toxic as Gesarol SH20 (loose emulsion) (Table 2). However, since constant agitation of the test solutions by aeration during the runs may have modified any specific degree of emulsification, it is probable that the values of toxicity as expressed by these products was not influenced by the nature of the emulsifying agent. Also, since the figures comparing the two Gesarols show a slight increase in toxicity in the presence of an emulsion, it is apparent that Surber's (loc. cit.) conclusions are correct.

The three products, two commercial (Neocid Larvacide and Neocid Barn Spray) and one of laboratory preparation (i.e. kerosene plus DDT) designed for control of insects affecting man, are probably of more importance from a fisheries biologist's standpoint than the products previously discussed, since the use of the latter products frequently involves the environment of fishes. All three products were found to be toxic to trout; and again, as with the former products, a considerable variation in specific toxicity existed. Kerosene plus DDT was the most toxic of them all, exhibiting a consistent tolerance limit of 0.06 p.p.m. DDT; and the wettable powder (Neocid Barn Spray) was the least toxic, its consistent tolerance limit standing at 10.0 p.p.m. DDT (Table 3). The two Neocid products, although exhibiting a marked contrast in their degree of toxicity to brook trout, did possess a striking similarity in the pattern of velocity of death. In concentrations of 50.0 p.p.m. DDT (Barn Spray) and 4.0 p.p.m. DDT (Larvacide) the minimum survival time was 24 and 25 hours, and the maximum survival time 87 and 95 hours, respectively. At 10.0 and 1.0 p.p.m. concentrations, the two substances, listed in the same order as above, were non-lethal in a 96-hour test. The velocity of death of the trout in the kerosene solution was somewhat modified in comparison to that in the Neocid products in which it was demonstrated that a sharp break occurred between lethal and non-lethal dosages; starting at a concentration of 0.8 p.p.m. DDT the minimum survival times was 2 hours, gradually extending to 44 hours at 0.08 p.p.m. DDT. The latter figure represents a concentration immediately above the one establishing the consistent tolerance limit.

The most striking contrast of these three products was the difference in toxicity between the wettable DDT powder (Neocid Barn Spray) and the

Table 3.--Hours survival of brook trout in different concentrations of oil base and inert powder DDT products designed specifically for use in control of insects affecting man and domestic animals.

Concentration of DDT (p.p.m.)	Number of test animals used for each product	Products Tested								
		Neocid larvacide			Neocid Barn Spray			Kerosene-DDT solution[1]		
		Hours survival			Hours survival			Hours survival		
		Minimum	Maximum	Average	Minimum	Maximum	Average	Minimum	Maximum	Average
50.0	10	...	...	...	24	87	51	...	...	...
40.0	10	...	...	...	24	67	48	...	...	...
35.0	10	...	...	...	43	91	69	...	...	...
30.0	10	...	...	...	44	63	50	...	...	...
25.0	10	...	...	...	48	96	86	...	...	...
20.0	10	...	...	...	48	96	75	...	...	...
15.0	10	...	...	...	43	96	77	...	...	...
10.0	10	...	...	...	96	96	96	...	...	...
8.0	10	...	...	...	96	96	96	...	...	...
6.4	10	...	...	...	96	96	96	...	...	...
4.0	10	25	95	64	96	96	96	...	...	...
2.0	10	25	96	62	96	96	96	...	...	...
1.0	10	96	96	96	96	96	96	...	...	...
0.8	10	91	96	95	96	96	96	2	49	33
0.6	10	68	96	87	96	96	96	2	64	42
0.4	10	87	96	93	...	...	...	16	60	39
0.2	10	96	96	96	...	...	...	40	64	47
0.1	10	96	96	96	...	...	...	...	...	...
0.08	10	96	96	96	...	...	...	44	88	60
0.04	10	96	96	96	...	...	...	96	96	96
0.00	10	96	96	96	96	96	96	96	96	96

[1] Prepared in test laboratory--not a commercial product.

oil base products (Neocid Larvacide and kerosene plus DDT). It is apparent that a considerable degree of the toxicity of the latter two products has been brought about by the method of compounding. Both contain petroleum products and likewise both constitute solutions, while the former product contains DDT and an inert material to act as a wetting agent and is insoluble in water. Undoubtedly the petroleum oils used as solvents and carriers in the former products themselves possess varying degrees of toxicity which materially added to the overall toxic values. However, it is believed that the major factor contributing to their increased toxicity is a result of the changes in the physical state of DDT in each of the two solutions. When in soluble form DDT would not only be more uniformly dispersed throughout the test medium, thus increasing the possibility of contact with the test animal, but would presumably also be present in a state whereby absorption would be greatly increased. The mode of entrance of powdered DDT to the test animal would be primarily by injection (Ellis 1944) or by contact with the gill filaments. In contrast, it is believed that DDT in solution could gain entrance to the host not only in the above manner but also through contact with the fin membranes or at any other exposed area of the body by absorption, as has been demonstrated with warm-blooded animals (Woodard, 1944).

Discussion

In order to make a direct comparison of the results of our studies with those of other workers the values derived in p.p.m. of DDT, which represent the maximum tolerance limit of brook trout, have been converted on a theoretical basis into pounds of DDT per acre-foot of water as follows:

Name of product	Consistent tolerance limit of brook trout (p.p.m. DDT)	Conversion to pounds of DDT per acre-foot of water
DDT plus kerosene	0.06	0.108
Gesarol SR20	0.10	0.27
Gesarol SRN20	0.20	0.54
Neocid Larvacide	0.20	0.54
Gesapon 18	0.60	1.63
Neocid Barn Spray	10.00	27.2

The first four products listed above, possessing similarity in composition (petroleum oil base), theoretically could be applied at rates varying from one-tenth to one-half pound of DDT per acre without any deleterious effect to trout. In comparing these figures with those derived by other workers who obtained their information through field and laboratory studies under a multiplicity of varied conditions, it can be noted that a certain degree of similarity exists. For example, Surber (1946) reported that an oil-base DDT product applied at the rate of 2.0 pounds per acre killed many species of fishes in the Patuxent Research Refuge, Bowie, Maryland. Also, in Ash Creek, Lackawanna County, Pennsylvania, a concentration of 0.23 pounds of DDT (oil base) per acre was sufficient to kill about 6 percent of the brook trout being held captive in the stream as test animals. In addition, Surber reported that many other species of fish were also killed in this stream. In an experiment involving the use of ponds, Surber found that brook trout were able to tolerate an application of 1.0 pound of DDT in kerosene per acre, while certain warm-water species were affected. Since a constant flow of water was continually entering the ponds, it is possible that the original DDT content became sufficiently diluted in a period which normally would be required to produce death, and hence the true value of the DDT concentration may actually have been much lower. In dealing with warm-water species in soft-water ponds, Surber found that applications of DDT at the rate of 0.10 pound per acre resulted in destruction of 43 percent of the total population of fishes present, and with applications of 1.0 pound per acre, severe loss (80 percent) of the fishes occurred. In hard-water ponds, applications of 1.0 pound of DDT per acre proved to be deleterious.

Reports issued by Cottam (1946) showed that concentrations of DDT (carried in an oil base) at the rate of 0.5 pound per acre killed warm-water fishes at the Savanna Ordnance Depot in Carroll County, Illinois, and at Island Beach, Ocean County, New Jersey.

Since the primary use of DDT is for the control of undesirable insects and the resulting fish kills are incidental to such applications, it is desirable to present at this time a few literature citations showing the concentrations of DDT customarily employed for this purpose so that the reader can determine for himself the relationship which exists between the two studies. Gerlach (1946) reported successful control of anopheline mosquitoes with concentrations of DDT in oil varying from 0.2 to 0.5 pound per acre; Hess (1947) indicated that successful kills of anopheline larvae were accomplished with DDT in a form of thermal aerosol at rates of 0.012 to 0.001 pound per acre; Hoffmann, et. al. (1946) cited instances in which applications of DDT in oil at rates varying from 0.23 pound per acre in Ash Creek, Pennsylvania to 1.0 pound per acre in Bowback Creek, Pennsylvania, were effective in controlling aquatic insects.

From the past citations covering data for both fishes and insects, it can be noted that the tolerance thresholds show a considerable amount of overlapping. DDT in very minute quantities was demonstrated as being toxic not only to insects but also to fishes. In glancing over the literature pertaining to this substance it appears that the least effect to fishes occurs when DDT concentrations are applied at rates less than 1.0 pound per acre. In comparison, four of the products used in these tests, similar in composition to those used in the field, were found to possess similar toxicity values.

Conclusions

1. In barely lethal concentrations of DDT products, brook trout did not show any signs of distress during the first few hours of exposure and generally it required a period of from 8 to 15 hours to produce death; and further the highest mortality occurred within the first 36 hours.

2. DDT products having a petroleum oil base were significantly more toxic than wettable DDT powder and DDT carried in a plant oil base. Theoretically, brook trout could be exposed to concentrations of DDT in petroleum oils ranging from 0.1 to 0.5 pound per acre without any deleterious effect. In the plant oil base DDT product, safe applications would be at the rate of 1.5 pounds per acre, and in the wettable powder, 27 pounds per acre.

3. Since the total toxicity to brook trout could not be accounted for in terms of DDT alone present in each of the products tested, it was concluded that the solvents and carrying agents employed in compounding contributed materially to the overall toxic values.

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