Field and Laboratory Studies of DDT and Aquatic Insects

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When DDT is put on woodlands to control defoliating insects, many creatures are affected besides the target insects. The forest lands of Connecticut are interlaced with streams that receive some DDT incident to aerial spray operations. The following review is limited to investigations of DDT in the fast moving waters of streams and their associated insects.

Literature Review

Many of the effects of DDT on aquatic life can be related to the physical characteristics of DDT and its relationship to objects within the water. DDT is only very slightly soluble, perhaps the least water-soluble of any known organic compound (Haller *in* Brown 1951). This solubility has been variously stated to be from 0.0002 parts per million (ppm) to 1.0 ppm. Bowman *et al.* (1960) found that if the minute undissolved particles formed in preparing saturated aqueous solutions were removed by ultra-centrifugation, the true solubility was not more than 0.0012 ppm at 25° C. One year earlier, Bowman *et al.* (1959) had confirmed and expanded upon the observations of earlier workers concerning the fate of DDT in aqueous suspensions. At 1.0 ppm about 20 per cent of the DDT had settled out in 48 hours, whereas at 0.01 ppm, virtually none settled out. Moreover, DDT was hydrophobic and tended to collect on solid surfaces such as the walls of a container. Within 2 minutes, 39 to 56 per cent of the DDT was distributed on the walls of the container. At the lower concentrations of 0.01 ppm the hydrophobic DDT tended to collect at the surface of the water and lower the surface tension. Furthermore, at these low concentrations the DDT volatilized with the concurrent vaporization of the water. At higher temperatures more DDT was lost from the water by volatilization than at lower temperatures.

When an aerial spray of DDT is applied, variable amounts of the insecticide reach the forest floor and streams. The actual amount depends on the wind, topography, and amount of leaf canopy (Maksymiuk 1963). Studies by Friend and Cooke in Connecticut (Turner 1963) showed that when applied at ½ pound per acre, DDT was recovered at a range of 0.01 to 0.24 pounds per acre under trees and 0.06 to 0.21 in the open (which is probably similar to the amount falling in the larger streams). Graham and Scott (1959), after an application of 1 pound, found from nothing to 1.2 pounds per acre in a coniferous forest with the usual range 0.01 to 0.2. Hoffmann and Surber (1948) found an average of 0.39 pounds per acre with a range of 0.0 to 1.1 pounds after a 1 pound per acre application in an area of open fields and deciduous forest.

The fate of the DDT once it reaches a stream depends in part on the formulation. The usual material for forest insect control is a mixture of DDT and oil. Undoubtedly, most of the DDT is retained in oil floating on the surface, does not reach the body of the water beneath, and is rapidly carried downstream. Graham and Scott (1959) found that surface samples initially showed a great deal more DDT than subsurface samples, but within 30 minutes the level was about equal. Savage (1949) reported that oil sprays were emulsified in rapids and broken water so that bottom-living invertebrates suffered more severely in fast water than in slow water where the oil floated undisturbed.

The DDT passes off rather quickly from the body of the water, decreasing from 1.35 ppm to 0.01 ppm within 2 hours, and disappearing in 24 to 32 hours (Graham and Scott 1958, 1959). Because of the relative insolubility and hydrophobic properties of DDT, it would be expected that little or no DDT would be washed into a stream. Rosen and Middleton (1959) stated that runoff of rain water from treated forest areas carried chlorinated hydrocarbons into streams, and Morgan and Kremer (1952) reported DDT in streams after rainfall.

In contrast, Graham and Scott (1959) reported negative findings. This probably represents the usual case. Breidenbach and Lichtenberg (1963) found DDT in 13 of 101 sampling stations scattered over the nation (7 of 19 in the far west, 0 of 3 in New England), but at these monitoring stations no effort was made to determine whether the insecticide came from sewage, agricultural runoff, forest spraying, or accidental contamination. Nicholson *et al.* (1964) never detected DDT in the water of a stream in a heavily treated agricultural area. They suggested that the absence of DDT was related to its low solubility. Further evidence on the likelihood of rain washing DDT into a stream can be derived from work done with DDT leaching into undisturbed soils. Woodwell (1961) determined that some of the ortho-para isomer leached into the B_2 horizon of a forest soil, but this must have occurred over a period of years, as the less persistent and more toxic para-para isomer was not found below the top few inches of soil. Moore (1964) in checking a test area that had been sprayed with 70 to 100 pounds of DDT 6 to 13 years earlier, found that the first inch of soil contained a larger proportion of the insecticide than the following 4 inches combined. Grzenda *et al.* (1964) studied DDT in a stream following an aerial woodland spray for insect control. When the entire watershed was sprayed, the greatest amount was found immediately after spraying, with decreasing amounts for 2 months thereafter. However, as the carbon adsorption columns used in this investigation became clogged with detritus, it is possible that the DDT was on the detritus rather than in the water. The following year wool filters were used before the carbon adsorption columns and no DDT was found in either column or filter. Only "spot" spraying was done this second year. This type of localized spraying is more typical of Connecticut conditions than are the broad area applications often used in the Canadian Maritimes or the western U.S.A.

It would seem unlikely that any great amount of DDT would be washed into a stream after the initial deposit was carried away by the current, unless a rain was heavy enough to erode soil and litter into the stream.*

To summarize, if DDT is applied over a woodland stream in the usual oil mixture, only a portion, perhaps 20 to 40 per cent will reach the surface of the stream. Most of this will remain on the surface except where rapids and churning water work the insecticide into the body of the water. There is little settling out of the DDT and only extremely small amounts are dissolved into the water. Furthermore, the hydrophobic properties of DDT make it cling to almost any object it encounters in the stream. Consequently, because of stream flow, adsorption, and dilution, the amount of DDT in the water declines drastically in the first few hours after application.

What results do these effects have on the stream biota?

Simmons (1945) reported that DDT rapidly lost its toxicity to mosquitoes if mud was present in a test dish. Other workers reported decreased fish kill in muddy streams when compared to clear streams. Arnason *et al.* (1951) reported that DDT would control blackflies 17 miles downstream with evidence of lethality for 90 miles. Fredeen *et al.* (1953) further determined that 0.1 ppm of DDT in a single 15-minute application practically eliminated blackfly larvae up to 115 miles downstream, especially if the water was turbid. Then Fredeen (1953) discovered that DDT was adsorbed on suspended solids in the stream. This explained the great toxicity to blackfly larvae. These larvae strain passing matter from the stream for food. The DDT, adsorbed on this material, would not affect fish or other animals in the water but would remain toxic to any animal that swallowed the suspended solids. Presumably, therefore, DDT should also be especially dangerous to other animals, such as the caddisworm *Hydropsyche,* that strain the passing water, but this aspect has not yet been fully investigated. Berck (1953) gained increased sensitivity in determining DDT in watercourses by extracting the insecticide from suspended solids in large volumes of water. Likewise, Weidhaas *et al.* (1961) found that the loss of DDT to soil in water dispersions was 78 per cent in 24 hours.

Because of the affinity of DDT for objects in water, one might expect that DDT would persist on the surface of aquatic plants. Hoffmann and Drooz (1953) found DDT in moss 1 month after spraying. Algae 24 days after spraying yielded almost 25 ppm. Graham and Scott (1959) found DDT on aquatic vegetation 2 months after spraying, as far as 10 miles below the sprayed area. Cope (1961) found DDT on *Potamogeton* 16 days after spraying. There may be some differential removal of the DDT from the water, depending on the organism or object within the water (Holden 1962).

There may be other secondary effects on the vegetation. Adams *et al.* (1949), Morgan and Kremer (1952), Webb and Macdonald (1958), and

* In agricultural or lawn areas where heavier dosages or different insecticides are applied, the danger of contamination of water supplies from surface runoff is correspondingly greater (e.g. Young and Nicholson 1951, Nicholson 1959 with DDT, and Tarzwell and Henderson 1957 with dieldrin).

Filteau (1959) all observed increased algal growth following a DDT spray, presumably because the reduced herbivorous insect population was not able to graze off the excess growth. An attempt was made in the spring of 1962 to measure quantitatively the growth of algae in sprayed areas of Connecticut using the method of Grzenda and Brehmer (1960). Because of vandalism and drought, the experiment was not completely successful, but preliminary results suggested that increased algal growth is not an inevitable result of spraying under Connecticut conditions.

The type of formulation also governs the types of insects that will be affected. Savage (1949) found that a DDT and oil spray was particularly dangerous to insects living on the surface of the water, whereas Hoffmann and Surber (1948) found that a wettable DDT formulation left surface insects relatively unaffected.

Aquatic insects vary, of course, in habitat, (e. g. Harrod 1964) and it would be expected that this would in part determine the extent of contact with DDT. Savage (1949) suggested that stream pool insects had less contact with DDT and oil than insects of the rapids. Weidhaas *et al.* (1960) found that mosquito larval mortality differed among species on the basis of where they fed, and Hitchcock (1960) suggested that differences in mortality between stoneflies might be associated with habitat.

The developmental stage of an insect also probably governs its contact with the insecticide. Scott (1961) suggested that the stonefly *Pteronarcys californica* was less susceptible to DDT because it was in the egg or early nymphal stage when the DDT was applied. Hoffmann and Surber (1945) found that the portion of a population of the caddisfly *Chimarra obscura* that had pupated survived a DDT spray, while those that were still larvae suffered high mortality. For similar results in Connecticut, see "Fall samples," page 18, and "Accidental deaths in streams," page 25.

As might be expected from the physical data, the immediate toxic effect of the DDT, except for feeders on suspended solids, does not generally extend far downstream. Adams *et al.* (1949) found that surface Hemiptera reappeared about one mile below the spray area. Insects representative of the bottom fauna were put in "live cars" by Graham and Scott (1958) and placed at various points within and without the sprayed areas. All insects in the sprayed area died within 24 hours but mortality decreased with increasing distance until all insects appeared normal 1½ miles downstream. Filteau (1959) found that mortality decreased greatly beyond the sprayed area, probably due to dilution by unpolluted waters.

All observers agree that the greatest kill of aquatic insects comes in the first few hours. The sharp decline in mortality shortly after the spray is presumably a result of the physical disappearance of DDT from the water and of the early death of the most susceptible members of the population. No stream has been completely denuded by a single spray. However, if a stream is sprayed several years in succession, there is a catastrophic loss in all orders of insects. Webb and Macdonald (1958) and Webb *et al.* (1959) checked streams that had been sprayed at various times over a period of 7

years. One stream that had been sprayed for 6 successive years finally produced no insects whatsoever in the immediate post-spray sample. The other streams with different spraying histories suffered severe losses also, but varied somewhat by stream. This reduction of the insect population resulted in a concomitant increase in "worms" (Annelida, Nemathelminthes and Platyhelminthes). With the limited data available, it would appear that the loss of insects was caused by a failure to recover rather than a decline in a set population. Several workers have recorded a resurgence of Simuliids, which are heavily hit by DDT, in the year following a spray. Furthermore, Ide (1957) noted a great number of small Chironomids in the months following a spray, and he ascribed this to multiple generations and lack of predators, rather than to a natural resistance in the population. Hitchcock (1960) suggested that there was a fortuitous loss of different genera in different streams. Years of successive spraying presumably prevent re-establishment and increase loss of genera that had previously escaped by chance.

Dead insects are carried downstream by the current resulting in large "drift" counts. The surface drift may range up to 110 times the normal drift (Scott 1961). The bottom drift may be even higher, up to almost 240 times normal (Hoffmann and Surber 1945).

Counts of the bottom fauna before and after spraying have also been made by other workers (e.g. Gorham 1961, Frey 1961, Bridges and Andrews 1961). Although many fine studies have been made, no general principles have emerged. Unfortunately, detail has sometimes been lost because of inadequate sampling methods. Quantitative results based either on weight or number from single samples have sometimes shown so much variation that no reliable conclusion may be drawn. Furthermore, some otherwise excellent studies have identified the insects only to the ordinal level, and it is difficult to determine what is actually happening in the stream at this taxonomic level. Consequently, one author has stated categorically that Ephemoroptera are the most susceptible of all aquatic larvae. Other authors have stated that Trichoptera are most affected. Undoubtedly each is true in a particular area, but it would help if a further breakdown to genera or species were possible so that meaningful comparisons could be made. When, in addition to this, differences among streams are considered, it becomes extremely difficult to compare studies in various parts of North America. Generally DDT has appeared to be most toxic to the Trichoptera and Plecoptera.

DDT persists for some time on the vegetation above a stream. Two weeks after a 1 pound per acre aerial application was made in Connecticut, a leaf sample from the treated woodland showed 21 ppm DDT wet weight. What effect this had on adults of aquatic insects can only be conjectured. Grzenda *et al.* (1962) believed that partial destruction of the adult population from a different insecticide caused a decrease in the aquatic larval population. Graham and Scott (1958) and Scott (1961) had some evidence from sweep net collections that adult aquatic insects were reduced following a woodland DDT spray, and Hoffmann *et. al.* (1949) noted a reduction of certain adult forms following an aerial spray.

It has been clearly shown that DDT may accumulate in the food chain of lakes or on land, but it is unlikely that this would occur to the same degree in stony streams (sens. Hynes 1941). Dead animals and other organic matter bearing DDT are eventually carried out of the stream to a lake or pond so that a stream may gradually cleanse itself. Hoffmann and Drooz (1953) discovered that DDT on moss accumulated toward the mouth of a stream rather than the head. Graham and Scott (1959) found DDT on aquatic vegetation both within and several miles below a sprayed area. Two months later the amount of DDT on aquatic vegetation within the sprayed area had declined considerably, but vegetation 10 miles downstream still retained about the same amount of DDT. Both of these results strongly suggest a downstream movement of DDT even after the initial spray material is carried off. Furthermore, fish or frogs killed by DDT are easy prey for terrestial predators and scavengers, and some insecticide is thus removed from the stream to the land. Peredel'skiy and Bogotyrev (1959) believed that aquatic insects caused "bioradiological purification" of water courses by removing radioactivity from the water to the land upon adult emergence. A similar phenomenon probably also occurs with DDT. It would appear, therefore, that the initial toxicity of the DDT is more important to stream life than the fact of the persistence of DDT in soil or the food chain, as would be the case in ponds or on land.

One other possible source of DDT in a stream occurs in the fall. Leaves dropping or blowing from deciduous trees might carry some DDT into the stream. Several aquatic insects use leaves as food or shelter, but the effects, if any, of DDT from this source are unknown. Several studies have reported fish deaths from DDT in the fall but these have usually been ascribed to the release of DDT stored in the fat. Woodwell and Martin (1964) suggested that DDT in coniferous forests may persist in the tree canopy and ultimately be carried to the soil.

Recovery of the insect fauna after a DDT spray has usually been left to natural forces. However, at least two attempts have been made to restock a stream with insects (Kerswill *et al.* 1958, Kerswill pers. comm. 1959) but with unpublished results.

In Canada very large numbers of small mayflies suddenly appeared in a stream that had extremely low numbers of insects after being sprayed for 7 consecutive years (Webb *et al.* 1959). This recovery possibly indicated development of resistance to DDT. No other case is known of resistance appearing in stream insects decimated incidental to spraying for woodland defoliators.

Recovery from DDT induced kills has been stated to take from a few months to several years. Hastings *et al.* (1961) found that recovery was not complete even after 4 years. After several years of blackfly control, Jamnback and Eabry (1962) found little difference in total numbers of arthropods between treated and untreated streams, but did find a decline in two orders of insects. The streams checked by Webb *et al.* (1959) showed consistent individual differences in recovery from DDT. In all of these studies, recovery was determined only to the order of insect. Some particularly interesting studies of recovery were made by Ide (1957, 1961). He captured adult insects upon emergence by means of a cage trap and was thus able to determine the insect fauna to species. The greatest qualitative loss was in species of Trichoptera. Numbers of insects in the treated streams were in some instances greater than in the untreated streams but the volume was lower, thus indicating a species shift toward the smaller forms. In New Brunswick a small stream recovered qualitatively in 4 years but the fauna of two other streams had not fully recovered in 6 years. Studies of Hoffmann and Drooz (1953) at the generic level indicated that recovery was essentially complete qualitatively within 1 year in a Pennsylvania stream.

Principles of DDT Action on Stream Insects

It is difficult to derive any principles of universal applicability from the effects of DDT on the insect fauna of streams, but the following general points appear established.

1. The amount of DDT reaching a stream from an aerial application is variable and depends on the type of application, tree cover, wind, and topography.
2. The greatest kill occurs in the first few hours and in areas nearest the spray point, before adsorption, dilution, and current have removed the DDT.
3. Apart from any expression of normal resistance, the susceptibility of an individual insect is determined by the interaction of formulation (determining whether the DDT floats or sinks, and the likelihood of lateral movement), the type of stream flow (rapid broken water, sluggish with pools), and habitat of the insect (water surface, mud, rapids, pools, etc.).
4. Turbid water causes DDT to be less toxic to aquatic animals, except those using the suspended sediments as food.

Two additional points might be surmised from published data.

1. Apart from the initial loss of DDT, there is a later movement out of, as well as down the stream, that removes toxic material from the stream.
2. Unless sediments (or possibly leaves) are washed into the stream, probably only minute amounts of DDT will get into the stream after the initial spray.

This study attempts to answer the following questions. What are the exact levels of insect mortality, apart from habitat differences? What are the recovery rates for streams in the deciduous woodlands of the Northeast? Will larger numbers of samples, determined at taxonomic levels below order, change or add detail to the generally understood picture of death and recovery?

Methods

A perfect means of sampling stream fauna has yet to be invented. The most common method is by use of the Surber foot-square sampler as in the present study. With this tool a large number of samples are needed to obtain statistically accurate quantitative results (Needham and Usinger 1956). In the present stream studies, 10 foot-square samples taken at each measurement were considered to give a qualitative measure of the number of genera present if 5 specimens of a genus were captured. The reasons for this standard and a description of the streams have been given previously (Hitchcock 1960). To test the possibility that fewer samples might be adequate, the collection from Bethany I in the spring of 1960 was taken in two sub-samples of five foot-square samples each, and all larvae were determined to genera. The results were: 17 genera occurred in both subsamples, 12 genera in only 1 subsample but in numbers less than 5 specimens and had not been captured before, and 10 genera in only 1 subsample but had been captured in earlier collections. These last 10 genera represent stream recovery from DDT that conceivably would not have been recognized with only five foot-square samples. Almost one-third of the total numbers of genera that would be considered important to stream recovery therefore would have been missed.

Insect drift from upstream can rapidly repopulate a denuded stream (Mueller 1954, Waters 1964). Consequently an attempt was made to sample only streams which would be sprayed from their source to a point well below the sampling station. This would prevent a population of susceptible insects repopulating the stream by drift, giving the impression that a decimated population had recovered, and possibly leading to a faulty conclusion as to the natural susceptibility of the population. This was possible in only three of the four treated streams. A fifth stream was sampled as an untreated check. All streams were within 6 miles of one another.

To test stream insects in the laboratory, it is necessary to provide cool, moving, well oxygenated water. To meet these requirements punctured tubing was coiled on the bottom of a plastic box as shown in fig. 1. A piece of screening placed over the coiled tubing gave firm footing to the larvae and allowed air pumped into the tubing to pass through. This diffuse aeration was necessary as insects cling tightly to a single source of air, making examination difficult. Two hundred millilitres of distilled water was brought to 60° F. and aerated for one-half hour before the insects and the proper amount of DDT were added. Use of distilled water avoided possible differences that might occur in stream water of varying quality. All testing was done in a controlled temperature room held at 60° F. As almost all of the DDT in a sprayed stream has passed off within 24 hours, it was not considered necessary to continue the tests beyond 24 hours.

The test insecticide was a 1 per cent emulsifiable solution of p-p' DDT. This was measured out to give the proper dilution for each test. The control was treated with equivalent amounts of the solvent (xylene and Triton X-100) alone. The only animal that had to be discarded because of failure

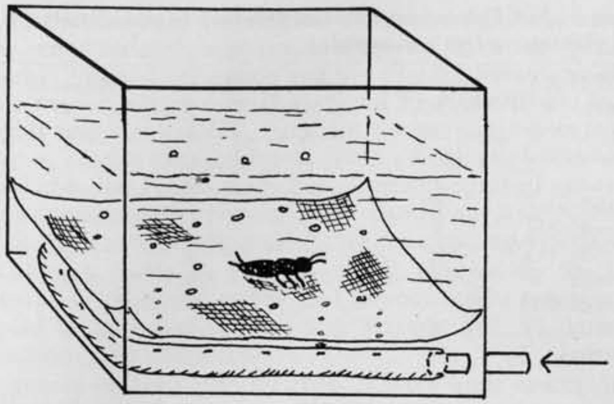

Figure 1. Punctured tubing coiled under screening in the testing box used in laboratory experiments provided for diffuse aeration while giving insect larvae a firm footing.

to survive was the amphipod *Gammarus*. Otherwise there was 100 per cent survival in the checks. At the higher levels with the most resistant insects, the resultant concentration of solvent caused difficulty, so a stock solution of 25 per cent DDT was diluted to give the required concentration.

As noted above, DDT will disappear from the body of the water to the walls of the container. Therefore, the insects in these tests were unavoidably subject to a decreasing amount of DDT over the test period. This has its counterpart, however, in a similar disappearance of DDT from water in nature.

It was necessary to test all insects of one species in a relatively short period of time because differences in size and age change the susceptibility of the insects to DDT.

Spring Field Tests

During the course of a routine airplane application of DDT at 1 pound per acre in the spring of 1959, it was possible to examine five streams before and after spraying, and in spring and fall for 2 years after the spray was applied. All treated streams but one (Bethany III) were sprayed from the headwaters to below the sampling point to eliminate downstream drift as a source of repopulation. Loss of insects in the field and subsequent recovery could thus be measured for a variety of genera in several streams over a couple of years.

The Ephemeroptera (table 1) proved to be relatively resistant to the effect of the spray. *Ameletus, Stenonema,* and *Baetis* appeared to be most affected, and these genera have also suffered heavy losses from DDT in other areas, according to the work of Hoffmann and Surber (1945), Hoffmann and Drooz (1953) and Ide (1956). With the exception of *Ameletus* in a single stream, all these had reappeared within 2 years. Although

Table 1. Survival of Ephemeroptera and Odonata in streams after a DDT spray (10 square feet per sample)

EPHEMEROPTERA	Hamden I	Bethany I	Bethany II	Bethany III	Cheshire check
Stenonema	X^1	X^1	X	X^1	X
Epeorus	X^1	X^1	P	X^1	X^1
Ameletus			O		
Baetis	P		X^1	X^2	X^1
Habrophlebia	X	X	X^1		X
Paraleptophlebia	X	X	X	X	X
Ephemerella (s.s.)	X	X	X	X	X
Ephemerella (Eurylophella)	X	X	X	X	X
Ephemerella (Drunella)	X^1		X	X	X
ODONATA					
Agrion		X		X	A
Lanthus	X	X	X	X^1	X
Boyeria		X	X		X
Cordulegaster	A	X	X	X^1	

A—present before spray in numbers less than five and not recovered again.
O—present before spray in numbers of five or more and not recovered again.
X—recovered 1 week after spraying in any number.
X^1—present before and recovered 1 year after spraying in any number.
X^2—present before and recovered 2 years after spraying in any number.
P—not present before but recovered within 2 years after spraying (immigration?).

Table 2. Survival of Trichoptera and Diptera in streams after a DDT spray (10 square feet per sample)

TRICHOPTERA	Hamden I	Bethany I	Bethany II	Bethany III	Cheshire check
Rhyacophila sp.	P	X	X^1		X^1
R. fuscula	X^1	O			X
Lepidostoma	X	X^1	X		X
Neophylax	X^1	X^1	X	X	P
Hydroptila	A	X^2	X		
Hydropsyche	P	X^1	X	P	X
Cheumatopsyche		O	A	A	
Diplectrona	A	P	P	P	X^1
Sortosa	P	P	P	X^1	P
DIPTERA					
Simuliidae	X^1	X^1	X^1	X^1	X^1
Chironomidae	X	X^1	X	X^1	X
Antocha	X^1	X^1	X^1	A	
Dicranota			X	X	X
Pseudolimnophila	X	X^1	X		X

Notations as in table 1.

Stenonema was found in the immediate postspray counts in Bethany II, generally the Ephemeroptera fared worse in this stream than in the others. Most of the missing mayflies had re-occurred by the fall of the first year in

all streams except this one. The *Ephemerella* were relatively unaffected and appeared to suffer little or no loss quantitatively from the spray. That this was a true physiological resistance and not due to conditions of its habitat was shown in the laboratory tests (figs. 8 and 9). Not only are some Ephemeroptera highly resistant to DDT, but there is some suggestion that resistance may develop in heavily sprayed forest areas (Webb *et al.* 1959). The presence of mayflies in the irrigation ditches of heavily sprayed agricultural areas of California (personal observation) may also be suggestive, but cannot be ascribed with assurance to acquired rather than natural resistance.

The Odonata (table 1) were also little affected by the DDT spray. Most Odonata are relatively sessile and in some cases remain semi-buried so they would be expected to have little contact with the insectide. Some natural resistance may also occur.

Some genera of Trichoptera (table 2) were quite susceptible to DDT. *Rhyacophila fuscula, Hydroptila,* and *Cheumatopsyche* were particularly slow to recover and had not returned to all streams even after 2 years. All streams did not respond in the same manner, and Bethany I appeared to recover more slowly than the others. Hoffmann and Drooz (1953) recorded *Rhyacophila* as relatively unaffected, but this undoubtedly represents a species difference. In table 2, *Rhyacophila sp.* was not affected so greatly as *R. fuscula.* Ide (1957) found only one species of Rhyacophilidae surviving in his treated streams although there were six or seven species in his check stream.

Specimens of *Diplectrona, Sortosa,* and *Wormaldia* appeared in several streams in the fall of the year after spraying and were found in all subsequent counts although not found in pretreatment counts. Two years after the spray, *Sortosa* was the most numerous trichopteran in Hamden I and *Diplectrona* the commonest one in Bethany II. This pattern did not appear so strikingly in the check stream nor in Bethany III (which received insect drift from an untreated headwaters). Another insect, *Hydropsyche,* was hard hit by the DDT spray but recovered quickly and unlike most of the caddisflies was found in the samples of the first fall. These particular caddisflies probably have the ability to recolonize rapidly. It would enable *Diplectrona, Wormaldia,* and *Sortosa* to colonize areas that might be otherwise unavailable to them before the other Trichoptera were reduced by the spray. As *Sortosa* appeared in the check stream for the first time in the year following the spray, some of this population shift may reflect a natural increase in numbers of this genus over the whole area, and this would also aid them in recolonization of the treated streams.

Overall, the Trichoptera were the most affected and the slowest to recover of all the aquatic orders. This varied, however, by genus.

The dipterous fauna in Connecticut appeared particularly susceptible to the DDT spray (fig. 3), but almost all genera had reappeared within a year (table 2). This included not only the smaller midges and blackflies but also the larger Tipulidae. The Simuliidae emerged as adults in the check stream between the time of the first two counts (table 2) so no direct measure of the spray could be made, but the effects of DDT on these insects and their subsequent resurgence has been well established by many previous studies.

The megalopterans (table 3) appeared relatively unaffected. Hoffmann and Surber (1945) reported similar results with *Nigronia* but Hoffmann and Drooz (1954) stated that the numbers of *Nigronia* were reduced, although their tables do not seem to bear this out. Because of reduced competition from other predacious forms, it might be expected that the numbers of *Nigronia* would increase in subsequent years. This increase took place, but 2 years later almost all the *Nigronia* captured were small ones.

The coleopterous psephenid *Ectopria* was eliminated by the spray but had recolonized the streams within a year (table 3). The elmid *Promoresia* was affected in the adult stage but larvae survived to preserve the population.

Table 3. Survival of Plecoptera, Coleoptera, and Megaloptera in streams after a DDT spray (10 square feet per sample)

	Hamden I	Bethany I	Bethany II	Bethany III	Cheshire check
PLECOPTERA					
Nemoura	X	X	X	X	X
Leuctra		X[1]	X[1]	X[1]	X
Peltoperla	X	X	X	X	X
Isogenus		X	X	X[2]	X
Isoperla	X[1]	X	X	X[1]	X[1]
Acroneuria	X[1]	O		A	P
COLEOPTERA					
Ectopria	X[1]	X[1]	X[1]	X[1]	X
Promoresia adult	X[1]			X[2]	X
Promoresia larva	X			X	X
Stenelmis	X				
Anchytarsus	X	X	X		
MEGALOPTERA					
Sialis		X[1]	X		
Nigronia	X	X	X	X	X

Notations as in table 1.

As might be expected, some genera of Plecoptera were more sensitive to the DDT spray than others (table 3). The phytophagous *Nemoura* and *Peltoperla* appeared in as great numbers immediately after the spray as before the spray was applied. They may have avoided contact with some of the DDT by their habit of remaining concealed in leaf packets. There was, however, a later reduction of the population of *Peltoperla* that was not immediately apparent in the postspray counts. This is discussed under "Fall samples" (page 18) and "Delayed effects of DDT" (page 23).

The carnivorous *Acroneuria* had not reappeared in two streams, even 2 years after the insecticide application. To a lesser extent, this reduction also occurred in the predacious *Isogenus, Isoperla,* and *Alloperla.* It appeared that the predacious stoneflies might be more susceptible than the

Table 4. Number of immature Plecoptera captured in samples of 10 square feet

	Spring pretreat.	Spring postreat.	Fall spray yr.	Spring 1st yr.	Fall 1st yr.	Spring 2nd yr.
			Hamden I			
Phytophagous	26	37	18	84	30	110
Predacious	7	0	10	5	11	11
			Bethany I			
Phytophagous	85	94	9	430	18	149
Predacious	25	18	16	18	45	18
			Bethany II			
Phytophagous	49	52	7	86	12	47
Predacious	11	16	3	7	8	15
			Bethany III			
Phytophagous	64	32	78	—	65	46
Predacious	37	0	11	—	50	17
			Cheshire check			
Phytophagous	51	9	78	31	13	5
Predacious	32	11	70	48	31	12

herbivorous forms and this facet was therefore examined more closely.

Table 4 shows this relationship. Predacious forms appeared to have been drastically reduced in two of the treated streams, but scarcely or not at all in the other two. The only pattern was one of irregularity between streams. This difficulty was compounded by the extreme variability of the check stream. A resurgence of phytophagous stoneflies (principally *Nemoura*) in Bethany I in the year following the spray cannot be attributed to the disappearance of predacious forms, as this was a stream in which carnivores, except *Acroneuria,* were unaffected.

However, if these spring data are graphed, it is notable that unlike the treated streams, both predacious and phytophagous forms in the check stream followed a similar population trend. The treated streams fell into two groups. Hamden I and Bethany III lost heavily in predacious forms and then recovered over the following 2 years, while the phytophagous forms remained constant or rose slightly. Bethany I and II did not lose many predacious stoneflies but the phytophagous forms rose in number in the first year after spraying and then subsided somewhat the year following.

No conclusion could be drawn except that differences between streams obscure the effect of the DDT on the stream fauna. The significance of this will be discussed below.

There were scattered collections of other invertebrates, but the only animal that appeared in numbers over five for the 10 samples in more than one stream was *Planaria.* It was apparently unaffected by the spray.

Figure 2 shows a summation of recovery as measured by the number of genera present in the spring samples. The stippled area shows genera with five or more specimens captured per ten samples. Summation of previous years after treatment is used for the columns of first and second years after treatment, since a few of the genera were found one year after spraying that were not found the second year. Presumably they were present. The white areas in the first columns are the numbers of genera found in pre-spray counts in numbers less than five that were never found again. The white areas in the final columns are the summation of all genera found after the spray in numbers less than five that had not been captured in pre-spray samples.

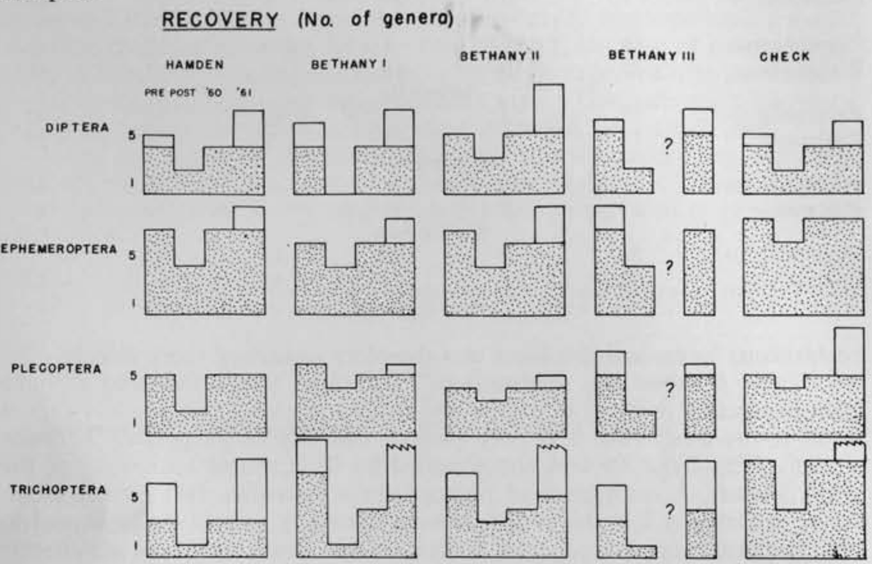

Figure 2. Number of genera of four orders found in spring samples (10 square feet per sample). Successive columns represent prespray, postspray, 1 year after spray, and 2 years after spray. Stippled area represents genera with five or more specimens per sample; clear area represents genera with less than five specimens per sample. Clear segment of last column represents summation of 1960 and 1961.

There was considerable variation between streams. For example, Bethany I lost all Diptera genera whereas Bethany II lost few. Hamden lost several Plecoptera genera while the other streams did not. The check stream also lost genera through emergence. The Trichoptera were most susceptible as individual genera, and even though the streams recovered with the same number of genera, the genera were not always the same as those originally there. The loss of genera within each stream was not generally a consistent loss. Every stream lost some genera but different streams lost different genera (table 1, 2, 3), and this would help speed repopulation as all streams were within 6 miles of one another. With a few exceptions all streams had recovered the number of genera equivalent to pre-spray counts within 2 years.

If we examine recovery as a function of total numbers, a different picture emerges. Figure 3 shows these relationships. It should be noted that the various orders are on different scales on the ordinate.

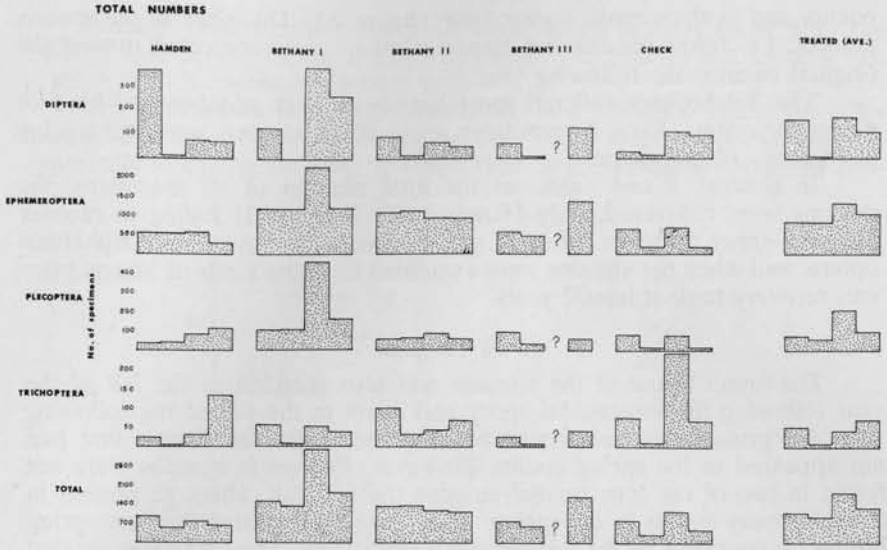

Figure 3. Total number of four orders found in spring samples (10 square feet per sample). Successive columns represent prespray, postspray, 1 year after spray, and 2 years after spray. Note differing scales on ordinate.

The Diptera suffered the most precipitous drop in numbers but recovered quickly in ensuing years. At Bethany I there was a strong resurgence in the numbers of Diptera (principally Simuliidae) following the spray. This pattern has been reported elsewhere several times. However, in this stream, this resurgence in number (figure 3) occurred in other orders, including the Ephemeroptera (which were little affected by the spray). This increase in Diptera, therefore probably had little relation to the spray in this particular instance. Hamden did not have this resurgence even though it had a high initial population of Diptera and indeed had not even reached its high initial level 2 years later. However, a qualitative comparison based on genera (figure 2) shows that Diptera exhibited the same approximate recovery pattern in both Hamden and Bethany I. That such increases are sometime normal, as might be expected, is indicated by the Trichoptera (principally *Hydropsyche*) of the check that increased 1 year after the spray.

The Ephemeroptera both qualitatively (figure 2) and quantitatively (figure 3) appeared relatively indifferent to the DDT, except in the aforementioned Bethany I and III. Bethany III, moreover, received insect drift from unsprayed areas upstream; other streams did not.

The dangers of depending on quantitative counts at the ordinal level to determine recovery is strikingly demonstrated with the Plecoptera at Hamden and Bethany I. Although the number of genera declined (figure 2), the number of specimens increased in both the immediate post-spray counts and in the counts 1 year later (figure 3). This shift in the species present, i.e. fewer species but larger numbers, was corrected toward the original balance the following year.

The Trichoptera suffered most heavily in both number and kind of any of the orders. Even 2 years later, some of the streams were still lagging in recovery of this order.

In general, if one examines the total number of all specimens, the streams soon recovered, only Hamden and Bethany II failing to recover their pre-spray numbers. Most of the specimens, however, were Ephemeroptera, and when the streams were examined as to the kinds of insects present, recovery took at least 2 years.

Fall Samples

The insect fauna of the streams was also sampled in the fall of the year following the insecticidal spray and again in the fall of the following year. No gross differences could be observed in the fall counts that had not appeared in the spring counts. However, *Peltoperla* nymphs were not found in two of the four treated streams the first fall, although present in the post-spray counts in the spring. They were also missing the next spring but had reappeared by the following fall and spring. The spray was applied before the adult *Peltoperla* emerged, but it was not possible to determine if the spray had any effect on emergence or on the adults.

The winter stonefly *Taeniopteryx maura* was found in all streams in the fall. The adults of this genus emerge and deposit eggs in the late winter or early spring. According to Frison (1929), the eggs of *T. maura* (=*nivalis*) hatch in the spring but the young nymphs show little growth before fall. It was surprising to find *Taeniopteryx* apparently unaffected by the DDT as smaller nymphs generally are more susceptible than the larger. The habits of the young *Taeniopteryx* are completely unknown. They may conceal themselves in the bottom rubble of the stream and so have little contact with the DDT in the water.

Laboratory Tests

Aquatic larvae were tested in the laboratory by the method previously described (page 10). Careful study was necessary to determine death-points for the various species. *Peltoperla* sometimes appear not to respond to a touch and to float helpless on the surface at 4 hours but will be apparently recovered at 24 hours. *Isonychia,* by contrast, is quite susceptible to DDT but will continue to quiver for many hours after it has passed the point of recovery. It thus becomes necessary to develop a scale of toxic effects for each individual species before regular tests can be made.

Generally the following three-unit scale was used: *alive-active,* coordinated movements or, in the case of sessile animals, the ability to right themselves quickly and accurately when turned over; *moribund-active,* uncoordinated movement or the inability, when swimming, to come to rest always in a normal position; *dead,* no movement to slight spasmodic movements. For the purpose of the dosage-mortality curves, mortality was considered to be *dead* only.

Initially, insects considered dead were placed in clear water after the 24-hour test period to see if any recovered. No species did so and this practice was discontinued in later tests.

The predacious stonefly, *Acroneuria,* is a common insect of riffles in brooks. I have observed it ranging over rocks and pebbles in mid-day apparently searching for prey. It would thus be presumed to come into con-

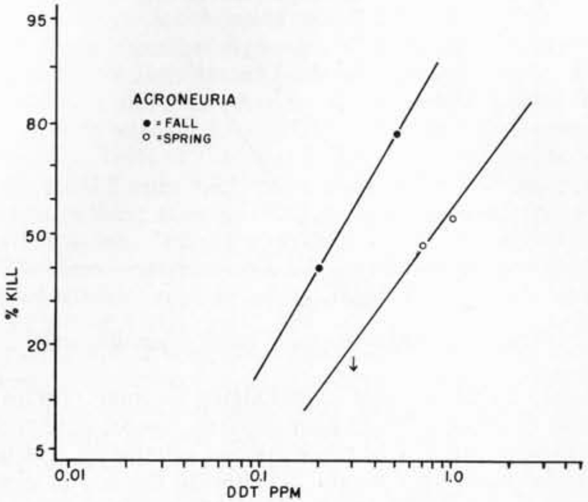

Figure 4. Dosage-mortality curve, log-probit scale.

tact with DDT in the water. Figure 4 shows the dosage-mortality curve for this insect in the laboratory tests. Jensen and Gaufin (1964) found *Acroneuria pacifica* to be relatively insensitive to O.1 ppm of DDT in water.

Paragnetina media, another predacious stonefly, appears to be closely similar to *Acroneuria* in the slope of the dosage-mortality curve. During the testing period it became obvious that the smaller specimens succumbed at lower dosages than the larger ones. Consequently, the test animals were arbitrarily broken into two groups–those insects 10-15 mm. in length and those 16 mm. or larger. These two groups were calculated separately. The results are shown in figure 5. The slopes of these two calculations are approximately parallel, but the greater susceptibility of the smaller larvae is shown by the LD_{50} at 0.33 ppm ($Y=4.10+1.74x$) whereas the larger larvae reach the LD_{50} at 0.66 ppm ($Y=3.32+2.05x$). The same pattern of parallel curves but greater susceptibility of the smaller larvae occurred in the *Acroneuria* when specimens from fall collections (small) were compared with spring collections (large).

Predacious caddisflies in the family Hydropsychidae were also tested and the susceptibility of these larvae proved to be in the same range as that of the two predacious stoneflies (fig. 6). LD_{50} was 0.78 ppm and $Y=3.18 + 2.34x$. At the conclusion of these tests, these *Hydropsyche*

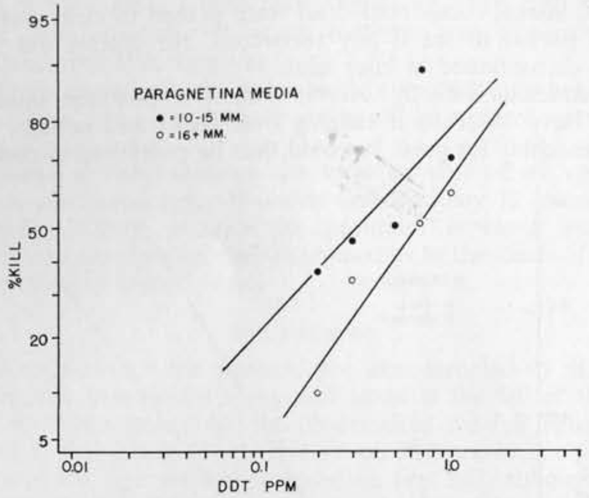

Figure 5. Dosage-mortality curve, log-probit scale.

were determined by O. S. Flint as belonging to three species–*H. betteni, H. sparna,* and *Hydropsyche* sp. near *morosa.* No breakdown was possible on the relative susceptibility of these species separately, but it may be assumed that individually they would fall close to the line as given.

A stonefly in a different niche, *Peltoperla maria,* was also tested. This insect ordinarily lives in leaf packets within the stream and feeds on dead

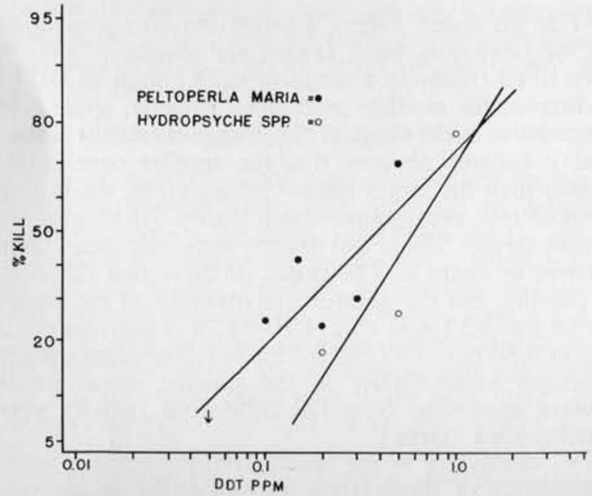

Figure 6. Dosage-mortality curve, log-probit scale.

leaves. This habit of concealment would presumably protect it in great measure from the immediate toxic effects of the insecticidal spray. The 24-hour LD_{50} for this larva is 0.38 ppm of DDT in the water ($Y=2.61+1.51x$), but the dosage-mortality curve is more flattened than that of *Hydropsyche* (fig. 6). The smaller fall specimens appeared to be more susceptible than the larger spring specimens, but a complete dosage series was not run on the fall specimens. A peculiarity exhibited by *P. maria* that did not appear with any other insect was the apparent recovery of larvae at 24 hours that were recorded as dead at 4 hours. This occurred only with spring larvae, not with those tested from fall collections. As this particular pattern of prostration and then recovery is sometimes typical of insecticide resistant insects, tests were made of DDT and WARF antiresistant for DDT compound (ARC). Tests of 0.1 ppm DDT plus 0.15 ppm ARC and also 0.2 ppm DDT plus 0.2 ppm ARC were made with 15 *P. maria* in each test. The mortality from these materials closely approximated that of equivalent amounts of DDT alone. The same apparent mortality at 4 hours and then recovery at 24 hours occurred with DDT plus ARC as with DDT alone. Therefore this particular mechanism of resistance was not one that could be overcome by use of ARC.

The apparent kill at 4 hours could conceivably mislead investigators who are making drift samples of dead insects in streams after spray operations. These *Peltoperla* would be carried away by the current, caught on screens, and recorded as dead but would recover if left alone. Of course, not all the *Peltoperla* captured in this manner would survive because predators and mechanical damage would kill some and others would fail to find a suitable habitat when they did recover. (See also "Delayed effects of DDT," page 23.)

A third important order of aquatic insects is the Ephemeroptera or mayflies. *Isonychia* sp. (det. B. D. Burks), a common larva, was tested with the results shown in figure 7. This insect appears to be quite sensitive to DDT and it was possible to make dosage-mortality curves for both 4 hours and 24 hours. The LD_{50} for 24 hours was 0.02 ppm DDT ($Y=4.09+2.42x$) and for 4 hours was 1.4 ppm DDT ($Y=2.00+1.39x$). Although these mayfly larvae were quite susceptible to DDT, they did not die quickly but continued to lie on their sides and move their appendages spasmodically for several hours. However, if these larvae were placed in clean oxygenated water, they did not recover, but invariably died.

Another mayfly, *Ephemerella subvaria* (det. B. D. Burks), proved to be the most resistant of any insect tested. The 24 hour LD_{50} was 17.31 ppm DDT ($Y=3.74+0.56x$). Moreover, the dosage-mortality curve was quite flattened (fig 8). It would seem likely that this insect would be able to survive quite high concentrations of DDT in a stream. The field data (table 1) provided support for this opinion as *Ephemerella* spp. were not obviously affected by the DDT that reached the stream.

A second species, *Ephemerella aurivillii,* was also tested. Figure 9 serves to illustrate differences within the genus. This large *Ephemerella* had a

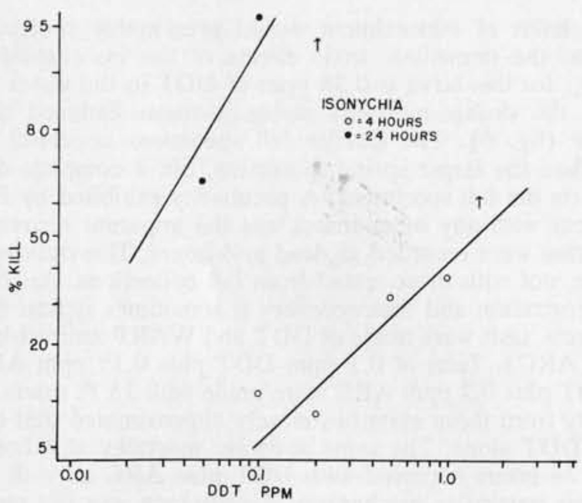

Figure 7. Dosage-mortality curve, log-probit scale.

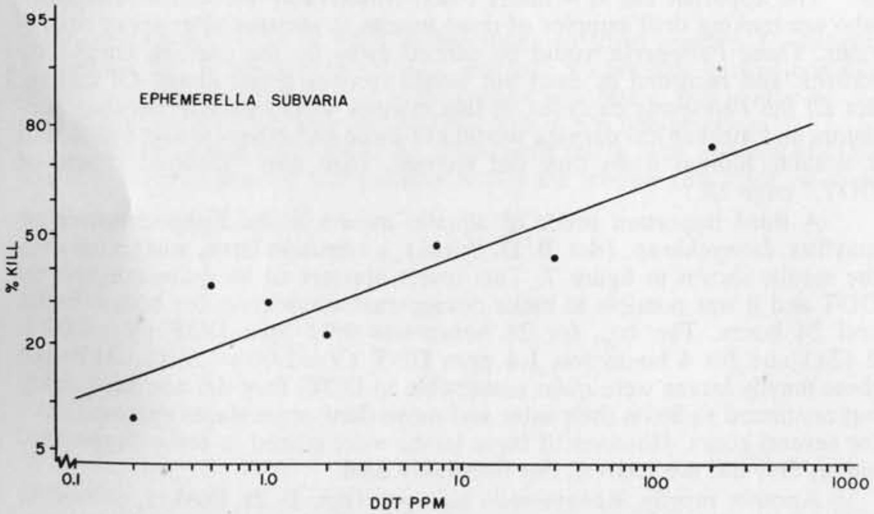

Figure 8. Dosage-mortality curve, log-probit scale.

24 hour LD_{50} of 4.8 ppm ($Y=2.55+1.45x$) and would appear to be more susceptible than *E. subvaria* to DDT.

In addition to the above insects, other insects were treated as time permitted, but not enough specimens were used to draw any definite conclusions. *Brachycentrus* nr. *numerosus* (det. O. S. Flint) was tested at 0.5,

1.0, and 1.5 ppm but the results were so erratic that no conclusions could be drawn. This caddisworm withdraws into its case at comparatively low dosages of DDT and this may possibly affect the amount of DDT actually reaching the animal. If removed to clean water it usually would resume movement. At times the larva would be found dead outside its case. Presumably it crawled out to die, rather than emerging from its case and being subsequently killed by the insecticide. The 24 hour LD_{50} is probably somewhere about 1.0 ppm.

Delayed Effects of DDT

Even though DDT soon disappears from the water it persists for some time on vegetation both in and out of the stream. Aerial DDT sprays are commonly applied in the spring and adult aquatic insects that emerge at that time would undoubtedly come into some contact with the residue remaining on leaves. Those insects emerging from small overgrown streams and those insects that remain hidden on the lower surfaces of terrestrial plants would

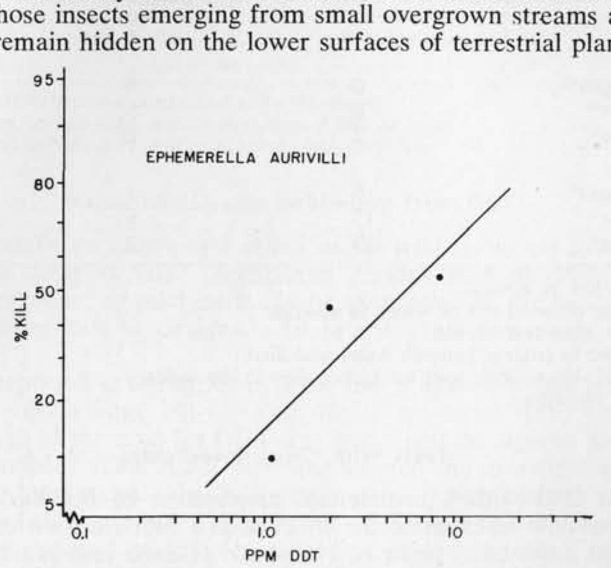

Figure 9. Dosage-mortality curve, log-probit scale.

undoubtedly contact less residual insecticide than those from larger, more open streams. However, no study has ever been made of the susceptibility of adult aquatic insects, nor of sources of DDT contact and the microenvironment, except on aquatic insects of medical importance.

During the latter part of the laboratory tests, it was noted that *Ephemerella subvaria* and *E. aurivillii* were emerging as subimagoes from the check but not from the DDT treated water. This was true despite the apparent indifference to DDT of the larvae when examined. The coordination, reactions, and abilities of the larvae appeared unimpaired, yet no emergence took place except in the control aquarium.

This was further checked by testing *Peltoperla maria* at the time of its normal emergence period. After *Peltoperla* had been in the insecticide for 24 hours, they were removed and placed in clear water with two leaves to see if any emergence would take place. The cumulative results are shown in table 5.

There appeared to be some delayed mortality and a differential emergence pattern in the *Peltoperla* treated with DDT. There did not appear to be any gross differences between those treated with DDT plus ARC and DDT alone. Almost nothing is known of the ecological requirements of *Peltoperla,* and undoubtedly the circumstances of this test were not ideal for the insect, but it seems equally clear that DDT may have more subtle effects on the individual insect than a test of immediate toxicity would show.

Table 5. Adult emergence of *Peltoperla* over a period of 6 days

	DDT	DDT + ARC	Check
Survived			
Emerged[a]	0	0	2
Larvae[b]	0	3	4
Larvae[c]	2	2	3
Died			
Emerged[d]	0	1	2
Larvae[e]	11	6	0
Larvae[f]	19	12	4

[a] emerged as adults.
[b] larvae crawled out of water to emerge.
[c] alive after test period.
[d] started to emerge beneath water and died.
[e] larval skin slightly split but larvae died in the water.
[f] died as larvae.

Tests With Other Insecticides

Field tests with a commercial preparation of *Bacillus thuringiensis* made it possible to examine the effect of this bacterium on stream insects. The results have been reported elsewhere (Doane and Hitchcock 1964). Laboratory tests on *E. subvaria* and *E. aurivillii* indicated that the bacillus had no effect on these insects. In the case of *E. aurivillii,* adult emergence took place in the aquarium treated with *B. thuringiensis,* as well as in the untreated aquarium.

A field test of Phosphamidon for forest insect control gave an opportunity to sample the stream insects that were incidentally under the spray. Results are shown in table 6. Using the same basis as that previously shown for DDT (Hitchcock 1960): 17 genera survived, 3 genera were eliminated and 5 genera possibly eliminated. The minimum loss of genera was 15.0 per cent and the maximum loss, 32.0 per cent. It would appear that Phosphamidon would present limited danger to aquatic insects.

Table 6. Survival of aquatic insect genera 5 days after an aerial spray of Phosphamidon

Ephemeroptera		Diptera	
Ephemerella (Eurylophella)	X	Chironomidae	X
Ephemerella s. s.	A	Simuliidae	X
Epeorus	X	Trichoptera	
Paraleptophlebia	X	Rhyacophila	X
Centroptilum	X	Diplectrona	X
Plecoptera		Chimarra?	X
Nemoura	X	Neophylax	X
Leuctra	X	Lepidostoma	X
Isoperla	X	Pycnopsyche	A
Alloperla	O	Psilotreta	O
Isogenus?	O	Psychomyiidae	X
Megaloptera		Hydroptila?	A
Chauliodes	X	Arachnida	
Sialis	X	Hydrachnellidae	A
Nigronia?	A		

X—present before treatment and after treatment.
O—present before only, and in numbers of five or more.
A—present before only, but in numbers less than five.

Accidental Deaths In Streams From DDT

Occasionally an excessive number of the wild fauna are killed following an aerial spray of DDT. Sometimes reasons such as double dosage, wrong turning point, or pilot error can be pinpointed as the cause. In other cases no reason can be assigned. In recent years two such kills were investigated.

It was reported in the spring of 1961 that Rimmon Brook in Seymour, Connecticut, had a large kill of trout due to an aerial spray of DDT. A routine analysis of the trout for DDT was made, and the aquatic insect population was sampled. This insect sampling assured the investigators that the fish kill arose from a cause that had simultaneously attacked other members of the stream fauna and not a cause, such as disease, that attacked the fish alone. In five square-foot samples, only five genera of insects were found (one of these was in the pupal stage). The only one represented by more than one specimen was *Ephemerella* s.s. (13 specimens), the most resistant of all insects tested in the laboratory. When compared with the insect fauna of the test streams, it was apparent that the brook had received a heavier application of DDT than normal.

A similar kill in Granby in 1962 showed similar results. Again there was a small larval insect population but one that included *Ephemerella.* However, various insects were alive on the banks of the stream. The habits of some of these insects indicated why they might be in an area of heavy DDT application (as verified by foliage and dead wildlife analysis). The mosquitoes *Aedes stimulans* and *A. abserratus* (det. R. C. Wallis) are spring snow-pool mosquitoes that would be sensitive to DDT but that could also migrate into the area. Generally caddisflies are rather sensitive to DDT but adult specimens of *Psilotreta frontalis, Phylocentropus lucidus,* and *Rhyacophila vibox* (det. O. S. Flint) were found. Dr. Flint stated that "these could have been, and probably were, pupae on May 18" at the time of spray application.

Discussion

It is apparent that recovery of the stream insect fauna is dependent on factors that have not yet been completely elucidated. Savage (1949) estimated that a stream would have regained its normal fauna in about 2 years whereas Ide (1961) found 4 to 6 years was necessary. In this study, the fauna had almost completely recovered (by a qualitative measure) after 2 years with only a very few genera missing that were represented in the prespray sample. Undoubtedly the size of the area sprayed is important in determining the speed of recovery.

As demonstrated in table 4, differences between streams are sometimes greater than differences between sprayed and unsprayed areas, although it is abundantly clear that there is a general reduction of insects after a woodland spray. The reasons for this variability remain obscure. In table 2, it may be seen that Trichoptera were qualitatively most affected in Bethany I whereas the Plecoptera and Ephemeroptera were hardest hit in Bethany III. Some insects, such as *Ephemerella,* are naturally resistant and survive in all streams, but with others there appears to be a fortuitous loss that may be dependent not only on the insect itself but also on the stream. Differences in topography, water volume and velocity, type of stream bottom, and overhead cover all regulate the amount of DDT which is available to an insect. As this varies from stream to stream, some types of insects are favored in one stream, whereas other insects are favored in a nearby stream. Bowman *et al.* (1959) indicated that DDT is hydrophobic and tends to concentrate on the upper water surfaces and the walls of a container. This strongly suggests that the habitat of an aquatic insect governs the amount of DDT that it will encounter. In addition, susceptibility to DDT varies among species. Webb *et al.* (1959) have noted in Canada also that the faunal response to DDT is not consistent between streams. This is fortunate as re-establishment of the insect population will be much quicker if there can be an interchange between streams.

In the present study, where a species was eliminated from one stream, there were survivors in nearby streams. Furthermore, because relatively small areas are sprayed in Connecticut, untouched refugia are left from which a population might expand to the treated portions of the watershed. If the untreated portion is upstream, insect drift will repopulate the sprayed portions quite rapidly. Little is known about the upstream movement of larvae, but gravid trichopterans will fly upstream (Roos 1957) and at least some species of both plecopterans (Frison 1935) and ephemerids (Burks 1953) can fly many miles under some conditions.

Most of the reports of large aquatic wildlife kills from forest insect spraying have been reported from coniferous rather than deciduous forests. This may be a result of more widespread spraying in the former areas or there may be other factors involved. Because of the adsorption of DDT on finely divided particles, it is possible that leaves falling into the stream decompose into detritus and tie up the DDT more readily in some forest types than in others. Although considerable work has been done on the breakdown of leaf litter on the forest floor, apparently little is known of the breakdown of leaves in a stream. Nykvist (1963) has pointed out that water-soluble substances are more easily leached from deciduous than from coniferous litter, but whether this might presage more rapid decomposition in a stream is apparently not known.

Winter is a particularly crucial time for fish in a stream denuded of insects by insecticides. Terrestial insects are available to a certain extent as food during the summer months, but during the cold months the fish are dependent on what is in the stream itself. Trout feed actively during the winter even at temperatures near freezing (Needham and Jones 1959) and must consume a considerable number of insects during that period. In this regard, the species of insect is important because only those insects which are large enough to be taken by trout will serve as food over the winter. Those insect species which overwinter as eggs or first instar larvae will not be available to the majority of the fish. Consequently, if the DDT had killed off most of those genera which served as winter food, one might expect a much higher fish mortality than if the DDT had killed those genera which overwintered in the early instars. Survival by genera as shown in tables 1 to 3 indicates that a broad enough spectrum of insect types can survive in each stream to provide a food source over the winter months. Results from late fall stream insect samples bear this out.

If the value of aquatic insects is considered to be principally that of a food source for game fish, then the effect of a spray lethal to wildlife is, for our purposes, of more importance to the insect population than it is to the fish population. Connecticut maintains an active fish-stocking program to satisfy the needs of sportsmen, and any game fish that might be killed could be replaced. However, there is no equivalent stocking program for aquatic insects in this country, although such a program has been suggested in Great Britain and has been tried on a small scale in Canada (Kerswill, Elson, and Keenleyside 1958). On this basis, a qualitative loss of insects would be more important than a quantitative one. If all insect species survived the insecticidal application, even at such low population levels that fish starved, density dependent factors presumably would come into play and the insects would survive.

The DDT from an aerial spray is diluted as it strikes the stream. Dissolved in oil, much of the DDT remains on the surface. However, the DDT in the water bathes exposed insects with a gradually decreasing amount over some hours. Generally the larger nymphs are more resistant than small ones of the same species, and thus the timing of the spray may also affect the survival rate.

The effect of DDT in a stream cannot be compared with that of a pond or marsh for the biota of a stream probably does not accumulate DDT to the same degree that the pond biota does. The movement downstream of DDT, and DDT bearing fauna, makes it less likely that DDT will get in the food chain.

The use of aerial DDT sprays over woodland streams in Connecticut destroys considerable numbers of aquatic insects. Fortunately, the infrequency of the sprays and the limited areas to which they are applied allows the stream insect fauna to recover within about 2 years. An incipient shift in the kinds of species within the stream is reversed as immigration restores the normal insect populations.

Summary

The insect fauna of four treated streams and one untreated stream was sampled 1 week before an aerial DDT spray of 1 pound per acre, 1 week after, and at subsequent intervals of 5,12,17, and 24 months. Ten square-foot samples at each time and location were considered to give an adequate qualitative representation of the number of genera present.

There was an initial heavy loss in the treated streams but the genera lost varied between streams. With a few exceptions the original insect genera had recovered qualitatively 2 years later. When examined as to total numbers, the pattern of recovery varied between streams and between orders. Two treated streams showed heavy loss and rapid recovery in numbers; the other two smaller loss but slower recovery. Generally it appeared that there were fewer species with greater numbers after the spray.

Laboratory tests in the three main orders of insects showed considerable specific differences in their responses to various dosages of DDT. Dosage-mortality curves were drawn for selected insects in the Plecoptera, Trichoptera, and Ephemeroptera. *Ephemerella subvaria* proved to be the most hardy of the insects tested with a 24 hour LD_{50} of 17.3 ppm. The stonefly *Peltoperla maria* was the only insect to exhibit recovery from the DDT dose. It appeared dead at 4 hours but had recovered by 24 hours. Younger insects were more susceptible than the older ones. There was some suggestion that DDT in the water affected adult emergence. The results on aquatic insects of an aerial spray with Phosphamidon is given.

Results of two field investigations of wildlife kills are given.

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