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Effect of Ultra ULV Fenitrothion Spraying on Brain Cholinesterase Activity in Forest Songbirds

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The organophosphorus insecticide fenitrothion (0,0-dimethyl-0-[3-methyl-4-nitrophenyl] phosphorothioate) has been used extensively for aerial control of spruce budworm (Choristoneura fumiferana) in New Brunswick. In recent years it has been formulated primarily as an emulsion utilizing a solvent, an emulsifier, and water as the carrier. A formulation consisting of a solvent and oil as the carrier has also been widely used. In both cases the active ingredient (AI) dosage has been 210 g/ha and the formulation application rate 1.46 L/ha. Boom and nozzle spray equipment has been standard.

A new formulation of fenitrothion is currently being tested for use against spruce budworm. Termed Ultra Ultra Low Volume (UULV), it consists of only two ingredients, 40% technical fenitrothion and 60% Dowanol TPM, and is applied at a rate of only 0.4 L/ha and the conventional dosage of 210 g AI/ha. The product is expected to work more efficiently with rotary atomizers, thus enhancing efficacy, and to lower operational costs because of the substantial reduction in materials required to be lifted to the target zone.

The effects of those formulations of fenitrothion on forest songbirds have been assessed previously by utilizing various evaluation criteria (e.g. Busby et al. 1983a, Busby et al. 1981, Pearce et al. 1979, Pearce et al. 1976). In general, results indicated that the margin of safety of fenitrothion is narrow and circumstances resulting in overspraying would have serious consequences for songbirds (Busby et al. 1983a). In other studies, it has been found that factors such as specific aircraft type, formulation, timing of spraying, and spray equipment used may bear on the environmental impact (respectively, Busby et al. 1983b and Pearce et al. 1979, Busby et al. 1983b and Pearce et al. 1979, Pearce 1975, and Busby et al. 1981).

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In view of those findings, the present study was conducted to assess the response of selected forest songbird species to aerial UULV fenitrothion spraying. Evaluation was based on measurement of brain cholinesterase (ChE) depression in wild birds exposed in the target zone.

MATERIALS AND METHODS

The study area was situated in the Maritime Lowlands Ecoregion (Loucks 1962) of eastern Canada, 18 km north of Doaktown, New Brunswick. Forest cover in the area was characterized by balsam fir (Abies balsamea), red spruce (Picea rubens), black spruce (P. mariana), eastern hemlock (Tsuga canadensis), and eastern white pine (Pinus strobus). Red maple (Acer rubrum), wire birch (Betula populifolia), beech (Fagus grandifolia), and yellow birch (B. alleghaniensis) formed a less significant portion of the species composition.

Treatment of the trial block was according to the normal operational procedure for spruce budworm control which consists of two spray applications, approximately five days apart, targeted at the third and fourth larval instars of the budworm. Technical details of the spray applications are given in Table 1.

Table 1. Technical details of aerial UULV fenitrothion applications.

	Spray 1	Spray 2
Block size	608 ha	
Insecticide formulation	40%/60% fenitrothion/Dowanol TPM	
Dosage rate	210 g AI/ha	
Application rate	0.4 L/ha	
Date and time	31 May, 09:30	6 June,09:00
Aircraft	team of two Cessna 188	
Spray equipment	Micronair AU4000 rotary atomizer	
Aircraft guidance	Aerial flagging system(Flieger1964)	

All birds were collected by shooting (0.410 shotgun). Sampling was done on the day of spray (Day 1) and the day following the spray application (Day 2). The sampling period was 11:00 to 18:00 on Day 1 and 06:00

to 14:00 on Day 2. Only males were sampled. Most were singing individuals although an attempt was made to collect non-singing birds. Unsprayed control birds were collected in nearby forest 6 June and 13 June. Birds in which the brain received heavy damage during collection were not included in the ChE analyses.

Species to be sampled were chosen on the basis of abundance within the spray block and to represent different foraging heights within the forest. The Tennessee Warbler (Vermivora peregrina) is essentially an upper canopy forager, the Bay-breasted Warbler (Dendroica castanea) forages primarily in the middle stratum, the Magnolia Warbler (D. magnolia) feeds in the lower crown, and the White-throated Sparrow (Zonotrichia albicollis) is found mainly in the ground-to-low-crown area.

After collection, the whole brain of each bird was placed in an air-tight serum vial which was stored in liquid nitrogen (-196 C) until laboratory assays were conducted. Brains were stored a minimum of 38 and a maximum of 47 days. Assay procedures followed the colorimetric technique of Ellman et al. (1961) as modified by Hill and Fleming (1982).

RESULTS AND DISCUSSION

Most samples exposed to spraying had a lower mean brain ChE activity than their respective controls (Table 2). In only two of the samples, the Tennessee and Magnolia Warbler, both from the Spray 2, Day 2 collection, were ChE values significantly lower than control values (t-test, $P<0.05$). In general, the greatest impact was observed in birds collected on Day 2, after Spray 2; the lightest impact was in birds collected on Day 1, of Spray 1. As a group, the Tennessee Warbler, the upper canopy forager and singer, exhibited the highest degree of ChE inhibition.

The impact of the sprays on individual birds is given in Table 3. The Tennessee Warbler was the species with the greatest number of individuals with ChE activity more than 2 SD below that of the controls; White-throated Sparrows had the fewest. The largest number of individuals of all species exhibiting inhibition of more than 2 SD was from the Spray 2, Day 2 collection; the smallest number was from the Spray 1, Day 1 collection.

Only two birds, a Magnolia Warbler from the Spray 2, Day 2 collection, and a Bay-breasted Warbler from the Spray 2, Day 1 collection, exhibited ChE activity levels more than 50% below that of the respective

Table 2. Mean brain cholinesterase activity† in forest songbirds: a comparison of unsprayed control birds and those exposed to aerial UULV fenitrothion spraying.

Species	Controls	Exposed			
		Spray 1		Spray 2	
		Day 1	Day 2	Day 1	Day 2
Tennessee Warbler	28.3(2.8)|11|◆	29.6(3.8)|12| 5▼	26.7(2.7)|14| -6	26.6(4.0)|15| -6	23.4(3.2)|13|★ -17
Bay-breasted Warbler	27.2(2.2)|10|	25.9(4.9)|16| -5	26.1(2.4)|11| -4	26.2(5.9)|10| -4	25.5(4.0)|10| -6
Magnolia Warbler	29.1(2.1)|13|	29.0(2.6)|11| -0	28.3(4.4)|14| -3	28.4(3.7)|13| -2	25.7(6.5)|12| -12
White-throated Sparrow	28.5(3.2)|12|	28.6(4.3)|11| -0	28.0(3.5)|12| -2	28.3(3.8)|9| -1	26.2(3.7)|11| -8

†μmoles acetylthiocholine iodide hydrolyzed per min/g brain (wet weight).

◆Mean brain cholinesterase activity (SD) |n|.

★Underlined figures are significantly different from controls (t-test, $p<0.05$).

▼Percent change of mean brain cholinesterase activity from controls.

controls.

Table 3. Number of individual songbirds exhibiting brain ChE activity more than 2 SD below the mean control values.

Species	Spray 1 Day 1	Spray 1 Day 2	Spray 2 Day 1	Spray 2 Day 2	Total(%)
Tennessee Warbler	1	1	3	7	12 (22)
Magnolia Warbler	-	3	1	3	7 (14)
Bay-breasted Warbler	3	1	2	3	9 (19)
White-throated Sparrow	-	-	-	2	2 (5)
Total	4	5	6	15	30 (16)

This study indicates that aerial UULV fenitrothion spraying caused ChE inhibition in songbirds exposed to the insecticide. The amount of inhibition depended on the species of bird and the preferred location of feeding in the forest canopy. There is now good evidence that upper canopy foragers (and, presumably, nesters) are the most affected by aerial spraying (e.g. Moulding 1976, Zinkl et al. 1977, Pearce et al. 1979). The results of the present study and an earlier one (Busby et al. 1983b) indicated that the ground-to-low-crown dwelling White-throated Sparrow is less vulnerable to forest spraying than the upper canopy birds.

Predictions of impact based on foraging height suggest that Bay-breasted Warblers would be more affected than Magnolia Warblers, contrary to the results of this study. However, the Bay-breasted Warbler is larger and of greater mass than the Magnolia Warbler (Dunning 1984), perhaps reducing its sensitivity. Furthermore, the foraging zones of the two species overlap considerably.

Our results suggest a cumulative impact each day following a spray application, as was found by Niethammer and Baskett (1983). Failure of the birds exposed in the first application to recover fully prior to the second spray is suggested, as ChE inhibition was slightly less in birds from the Spray 1, Day 1 collection than those from the Spray 2, Day 1 collection. Those results are consistent with the findings of Busby et al. (1983a) and Hamilton et al. 1981. Organophosphorus insecticides induce depression of ChE, which recovers primarily by de novo synthesis

(Fleming 1981), a relatively slow process varying with the species and insecticide involved. The present study indicated that the recovery period for forest songbirds exposed to UULV fenitrothion spraying is greater than the conventional five-day interval between spruce budworm spray applications.

Although significant ChE inhibition was detected, it is unlikely that mortality of birds occurred. In many studies it has been found that ChE depression greater than 50% is necessary to cause direct mortality (e.g. Bunyan et al. 1968, Ludke et al. 1975, White et al. 1982, White et al. 1983), although that may depend on the particular circumstances of exposure (Zinkl et al. 1980) and the species involved (Fleming and Grue 1981, Niethammer and Baskett 1983). In the present study only two birds had ChE depression of more than 50%, and in spite of more than 180 man-hours of time being spent in the block no signs of poisoning were observed.

The effects of sub-lethal ChE inhibition on the behavior, reproductive success, and survival of songbirds exposed to insecticides is poorly understood. Observations of reduced singing activity, nest desertion, and decreased nestling growth in White-throated Sparrows exposed to conventional fenitrothion spraying, but at double the normal dosage (Busby-unpublished data) indicated the potential for sub-lethal effects to be biologically significant. The present study suggests that UULV fenitrothion spraying may have slightly less impact on songbirds than conventional fenitrothion spraying. However because of the narrow margin of safety of fenitrothion, the UULV approach may cause substantial impact when overspraying occurs. Nestlings have been found to be especially sensitive to ChE-inhibiting insecticides (Grue and Hunter 1984) and should be further studied under field conditions.

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