

EFFICACY OF *BACILLUS THURINGIENSIS* VAR. *ISRAELENSIS* DE BARJAC FOR MOSQUITO CONTROL IN A WESTERN OREGON LOG POND¹

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ABSTRACT. Field tests of *Bacillus thuringiensis* var. *israelensis* were conducted in a moderately to heavily polluted log pond in Lane Co., Oregon containing a mixed population of *Culex peus* and *Cx. pipiens*. Five dosages were applied ranging from 0.40 to 1.63 kg/ha. Reductions of 73–99% were observed after 48 hours, but with no significant differences

among the dosage rates tested. One week following each test, treated areas had returned to pre-treatment levels of larvae, but with significant differences in age class distributions. Laboratory bioassays produced estimated ED₅₀'s approximately three times higher in water from the log pond compared with those conducted with distilled water.

INTRODUCTION

Bacillus thuringiensis variety *israelensis* de Barjac (*Bti*) is a promising new material for the control of larval mosquitoes (de Barjac 1978). It is highly toxic to some aquatic Diptera, especially mosquitoes and black flies (Garcia and Desrochers 1979, Undeen and Colbo 1980), but non-toxic to most non-target organisms, including other aquatic insects, other aquatic invertebrates, and vertebrates.

Although *Bti* has been tested in a number of aquatic habitats, including standing ponds, we are unaware of its being tested in log ponds. Log ponds have long been recognized as presenting unusual difficulties in mosquito control because of high organic content of the water and the relative difficulty of access to spraying equipment (Hoffman and Yates 1956, Ogden et al. 1960). In Lane County, heavy pressure from citizens against the use of conventional insecticides has made mosquito control even more challenging. Because of these factors we conducted a cooperative field test involving the Department of Entomology at Oregon State University and the Lane County Department of Health and Social Services.

MATERIALS AND METHODS

The material used in these tests was a wettable powder formulation, ABG-6108 supplied by Abbott Laboratories, *Bacillus thuringiensis* var. *israelensis*, Serotype H-14, and labeled as containing 2000 International *Aedes aegypti* toxic units per milligram. Laboratory bioassays were conducted before the field trial, using an adaptation of the method described by Sun et al. (1980). Data were analyzed by probit analysis using a program written in BASIC and run on a TRS-80[™] microcomputer. Because of suggestions that water quality could affect the effectiveness of *Bti* (Ignoffo et al. 1981), tests were run in both distilled water and log pond water. The field tests were conducted in a log pond in Junction City, Lane County, OR. The pond was approximately 2 ha (5 acres) in size, and was dissected into two approximately equal areas by an earth dam used to allow equipment access to the pond. Water depth was ca 1–2 m throughout the pond. Treatment was confined to an area measuring 61 × 91 m (0.56 ha [1.37 acre]) on one side of the dam; a similar area on the other side served as a control.

Although the treated and control areas were not completely isolated from each other, the connection between the two halves of the pond was at the opposite

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side of the pond from the study areas. The amount of *Bti* reaching the control area should have been negligible. As a standard method of estimating larval density, five 0.47 liter (1-pint) dips were made randomly at 4 different locations in both the treated and control areas. Sampling was done immediately before treatment, 24 hours, 48 hours, and one week after treatment. Samples were separated into age classes, but not by species. A separate sample was taken to estimate species composition. This sample consisted of 62% *Culex peus* Speiser, 38% *Cx. pipiens* Linn. Treatment was by a truck-mounted 379-liter (100-gal) capacity sprayer having a rotary agitator. Between July and September 1981 we conducted 5 experimental treatments. Dosage rates varied from 1.63 kg/ha (1.45 lbs/acre) to 0.40 kg/ha (0.36 lbs/acre). Application rates varied from 338.88 liters/ha (36.23 g/acre) to 899.34 liters/ha (96.15 g/acre). The weather was warm (mid-day highs ca 30°C) and clear during the entire test period.

RESULTS

A summary of overall results is shown in Table 1. There were reductions in the treated populations of from 73 to 99% after 48 hours over the range of dosage rates tested. We achieved a 90% reduction even at the lowest dose tested (0.40 kg/ha). All 24-hour and 48-hour samples from treated areas differed significantly from controls by t-test ($p < 0.05$). There were no significant differences, however, among any of the dosages tested. In all

cases, overall larval density in treated areas did not differ from that in control areas after one week. There were significant differences in age class distributions, however, with early stages accounting for a much lower proportion of total larvae in the treated area vis-a-vis the control area after 24 hours (Table 2). One week after treatment, however, early larval stages predominated in the treated area.

In preliminary bioassays using larvae collected in the treated area, we obtained an estimated ED_{50} of 0.357 IU/ml in pond water; an estimated ED_{50} of 0.111 IU/ml in distilled water. This 3-fold difference between pond water and distilled water agrees with similar bioassays we conducted with larvae and water from a log pond in Philomath, OR.

DISCUSSION

These field tests suggest that *Bti* applied to log ponds at rates within the ranges tested will provide satisfactory control of common log pond mosquitoes. The dosage rates and application rates tested were, with a single exception, within the range of rates recommended by the supplier. The presence of *Cx. pipiens* in high numbers (38% of those sampled) indicates a pond which is moderately to heavily polluted² and thus likely to represent a situation providing a strong challenge to chemical control of mosquitoes. A study of the post-treatment age class distribution substantiates the fact that *Bti* is effective only against larval mosquitoes, and more toxic against early stage larvae than late. It also suggests that

Table 1. Overall reduction (percent) in density of mosquito larvae after treatment with *Bacillus thuringiensis* var. *israelensis* (*Bti*).

Test number	Dosage (kg/ha)	Application rate (liters/ha)	Percent reduction (from controls)		
			24-hrs	48-hrs	1 week
1	1.63	338.88	74.49	98.96	43.16
2	1.46	610.04	89.13	85.95	-0.53
3	0.81	677.76	92.72	97.75	16.85
4	0.54	899.34	84.81	72.82	-19.19
5	0.40	677.76	56.61	90.52	-14.22

Table 2. Mosquito larvae in various age classes after treatment with *Bacillus thuringiensis* var. *israelensis* (Bti).*

Stage	Pretreatment	Post treatment				Control
		24-hrs	48-hrs	1 week		
	A	B	B	C	A	
1st-stage larvae	3431 (26.9)	368 (9.3)	200 (15.8)	5880 (56.7)	3748 (39.6)	
2nd-stage larvae	2814 (22.0)	541 (13.6)	189 (15.0)	2647 (25.5)	2147 (22.7)	
3rd-stage larvae	2568 (20.1)	952 (23.9)	209 (16.5)	999 (9.6)	1654 (17.5)	
4th-stage larvae	2456 (19.2)	1020 (25.6)	198 (15.7)	717 (6.9)	875 (9.3)	
Pupae	1505 (11.8)	1097 (27.6)	468 (37.0)	124 (1.2)	1071 (11.3)	
Totals	12774 (100)	3978 (100)	1264 (100)	10367 (100)	9455 (100)	

* Number of larvae followed by percentage (in parentheses) of all larvae in given treatment group. Combined data for 5 treatments. Columns differing significantly ($p = .05$) by χ^2 test are designated by differing letters at head.

reinfestation occurs quickly so that with the species of mosquitoes involved and the summer temperatures common in western Oregon, weekly treatment during the mosquito breeding season is necessary to achieve continuous control. Reinfestation was probably accelerated because we did not treat the entire pond, but the experience of others (Lewis and Eddy 1959) indicates that reinfestation occurs rapidly even when entire ponds are treated. It is thus doubtful that the treatment interval could be extended much. Even at the highest dosage rate tested (1.63 kg/ha) we were unable to detect evidence of toxicity after one week. Among the various dosage rates we tested, the only significant difference in reduction we noted was at the lowest rate (0.40 kg/ha) 24 hours after treatment, although the reduction was about 90% after 48 hours. We believe this is an indication that we had approached the lowest level of effectiveness—this dosage is, in fact, below the supplier's recommended minimum dosage (0.56 kg/ha) for polluted water.

The ecology of the log ponds is very complex, with the degree and type of pollution depending on a variety of factors—among many others; source of water, water flow, log storage time, species of logs, and size of pond.² Although previous investigators have reported apparent reduction in activity of insecticides

in log pond water from that in tap or distilled water (Hoffmann and Yates 1956), there is no specific information available concerning the mechanisms involved. Although the mosquito species present are considered indicative of moderate to heavy pollution, we were able to obtain a satisfactory level of control even at relatively light dosages of Bti. Other log ponds should be tested, however, to include ponds which vary in factors known to influence degree of pollution and mosquito species composition.

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MOSQUITO DISTRIBUTION AND ABUNDANCE IN AN INLAND SALT MARSH, SALTVILLE, VIRGINIA

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ABSTRACT. At an inland salt marsh, the most abundant mosquitoes collected were *Aedes sollicitans* and *Ae. vexans*. Their widespread occurrence was attributed to chloride tolerance ranges of 1,000-30,000 and 500-11,500 ppm, respectively. *Anopheles punctipennis* was also collected in pools containing chloride concentrations between 0.0 and 3,000 ppm. The

chloride concentration range of this species contributed to its association with *Ae. sollicitans*. Approximately 90-95% of the breeding sites of *Ae. sollicitans* and *Ae. vexans* were not located in areas where natural and historical landmarks occurred, therefore mosquito control procedures could be compatible with the preservation of this disjunct salt marsh.

INTRODUCTION

The presence of salt marsh mosquitoes at inland salt marsh sites has been reviewed by Felton (1944). The infestation of salt marsh mosquitoes in man-made salt-water habitats, such as roadside ditches, has recently caused concern among mosquito control workers (Berlin 1977). These man-made habitats have been infested not only by salt marsh mosquitoes, but also fresh-water mosquitoes (Kardatzke 1980). In Saltville, Virginia (81°52'N, 36°52'E) there exists a disjunct salt marsh (32 ha), 560 km from the eastern shore of Virginia. Several fresh-water and salt-water ponds and pools are present and range in salinity from 9-33,000 ppm. This salt marsh has been partially filled, drained and mined for salt several times in the past (Ogle 1981), but there

have always been numerous mosquito breeding sites present in the area.

Aedes sollicitans (Walker) is the most frequently encountered salt marsh mosquito at inland salt-water habitats (Felton 1944, Berlin 1977). Preliminary investigations indicate that it is also present in large numbers in Saltville. This mosquito is a major nuisance wherever it occurs, and in Saltville its biting habits caused an environmental conflict.

Not only is the area a favorite habitat of mosquitoes, but there are several attributes of natural and historical value which are considered worth preserving by several local groups. Five halophytic plants are present; *Atriplex patula* Linn. (orach), *Eleocharis parvula* (R and S) Link (spikerush), *Juncus gerardi* Loiseleus (black grass), *Salicornia europaea*