

FIELD EFFICACY OF VECTOBAC[®]-12AS AND VECTOBAC[®]-24AS AGAINST BLACK FLY LARVAE IN NEW BRUNSWICK STREAMS (DIPTERA: SIMULIIDAE)

C. M. RILEY¹ AND R. FUSCO²

ABSTRACT. The control of black fly larvae using 2 aqueous suspension formulations of *Bacillus thuringiensis*, serotype H-14 was evaluated in 6 trials on small and large sized streams in New Brunswick. Applications equivalent to 25 ppm (Vectobac[®]-12AS) or 12.5 ppm (Vectobac[®]-24AS) over a 1-min period resulted in 100% control of black fly larvae up to 3 km downstream of the application point. Over the 6 trials, effective carry was seen to increase with increased stream discharge which varied from as low as 0.8 m³/min in the small stream to 108.6 m³/min in the large stream.

INTRODUCTION

The development and use of *Bacillus thuringiensis* var. *israelensis* (*B.t.i.*) (serotype H-14) as a black fly larvicide has been reviewed by several authors (Gaugler and Finney 1982, Margalit and Dean 1985, Lacey and Undeen 1986). Field effectiveness of the early, wettable powder formulations of *B.t.i.* was limited by the large particle size which resulted in rapid settling out from suspension and poor "carry," or downstream transport, of larvicidal activity (Molloy et al. 1984). Despite the development of flowable concentrate formulations with reduced particle size and improved miscibility with water, effective larvicidal activity downstream of the application point has, with the exception of treatments made to rivers with very high discharge rates (Lacey et al. 1982), typically been measured in hundreds of meters (Lacey and Undeen 1984, Colbo and O'Brien 1984, Horosko and Noblet 1983, Pistrang and Burger 1984) compared with tens or hundreds of kilometers in some operations with chemical larvicides (Depner et al. 1980).

With increased public demand for target specific pesticides with reduced environmental impact, the need for formulations of *Bacillus thuringiensis* (*B.t.*) to be as economical and effective as chemical pesticides has been reduced. Although *B.t.* based pesticides are now regarded as an environmentally "safe" choice, the economics of large scale black fly control programs still remains an important consideration when deciding which pesticide formulation to use. In this regard, development and testing of higher potency *B.t.i.* formulations with improved efficacy and increased carry has continued (Lacey and Heitzman 1985).

The objective of this study was to provide

preregistration efficacy data for the use of 2 high potency *B.t.i.* formulations in eastern Canada.

MATERIALS AND METHODS

This study was conducted in 1987 on sections of Burpee Mill Stream and Bull Pasture Brook located within the Acadia Forestry Experimental Station, Fredericton, New Brunswick. The 2 formulations used were Vectobac[®]-12AS (ABG-6193, Abbott Laboratories) rated at 1,200 International Toxic Units (ITU)/mg and Vectobac[®]-24AS (ABG 6221) rated at 2,400 ITU/mg. Each product was tested on 3 separate occasions on sections of water located either upstream of these previously treated or 2-3 km downstream of previously unaffected stream sections.

The stream parameters for each of the 6 tests are listed in Table 1. Stream discharge rates were calculated immediately prior to each test using width, depth and stream velocity measurements. Depth was assessed at regular intervals to give at least 20 measurements across the width of the stream. Surface stream velocity was assessed over a 20-m distance for each treatment. Stream discharge was calculated by multiplying the average stream depth by the average stream velocity. Actual stream velocity was assumed to be 85% of the observed (surface) stream velocity (Horosko and Noblet 1983). Water temperature was recorded at the time of treatment.

Each test was comprised of 6 applications. Sections of shallow fast-flowing stream were sequentially dosed at 10-m intervals downstream from the previous application using either ABG 6193 at 1, 2, 3, 4, 5 and 10 ppm or ABG 6221 at 0.5, 1.0, 1.5, 2.0, 2.5 and 5 ppm maintained over 1-min periods. This procedure produced increasing cumulative doses at each successive downstream section for a maximum additive concentration of 25 ppm or 12.5 ppm, respectively. In each case, the appropriate amount of formulation was added to a watering can and diluted with stream water to give a total volume of 3 liters. The diluted formulation was mixed thoroughly and applied 10-20 cm beneath

¹ Chemical and Biotechnical Services, Research and Productivity Council, P.O. Box 20000, College Hill Road, Fredericton, New Brunswick, Canada, E3B 6C2.

² Abbott Laboratories, 4083 Rosewall Court, Harrisburg, PA 17112, USA.

Table 1. Stream parameters encountered during the evaluation of 2 Vectobac formulations for the control of black flies in New Brunswick during 1987.

Treatment	Location	Date	Vectobac formulation	Stream discharge (m ³ /min)	Width (m)	Water temp. (°C)	Comments
1	Lower Burpee millstream	July 1	12AS	108.6	17.0	16.5	No rocks exposed.
2	Mid-Burpee millstream	July 6	24AS	37.2	16.5	16	Rocks exposed, some moss.
3	Upper Bull pasture brook	July 10	12AS	0.84	2.0	17	Low water, moss-covered rocks exposed.
4	Mid-Bull pasture brook	July 27	24AS	1.26	2.4	17	Low water, moss-covered rocks exposed.
5	Extreme upper Burpee millstream	Aug. 4	12AS	18.6	5.1	16	Stream bottom covered with weed extending to water surface.
6	Mid-Burpee millstream	Aug. 6	24AS	13.8	5.0	16	Low water level, stony bottom with weed.

the surface of the water while walking steadily back and forth across the stream. The watering can was calibrated to empty after 1-min. The total application time for each test was 20–30 min. Stream beds were generally rocky and/or covered with weed; however, efforts were made to minimize the amount of silt disturbed.

Sequential collection of larvae from predetermined sample sites began at the upstream locations 1 h after treatment. Working downstream, subsequent samples were collected approximately 1 h after passage of the larvicide treatment. Sample sites were located at intervals between 20 m upstream (the control sampling site) and 4 km downstream of the first application point.

Black fly larvae were collected from artificial sampling surfaces which had been placed in the stream up to 10 days prior to treatment. Each artificial sampling surface was made from eight 20-cm plastic Ty-wrap® cable ties attached to a base of perforated steel angle approximately 30 cm long. Four such substrates were at each of the sampling sites. The colonized plastic Ty-wraps from each of the 4 artificial samplers were cut off and immediately placed in screw-topped, 1-quart mason jars containing ca. 750 ml of stream water. The jars (up to 4 from each sample site) were placed in coolers with iced stream water and returned to the laboratory.

In the laboratory, the lids were removed and the jars placed in a large tray (50 × 200 × 12 cm) containing 10 cm of cold running water for up to 48 h. Each sample was continually aerated with a gentle stream of filtered, compressed air supplied through an air stone.

The samples collected from each location were

divided into 2 groups and assessed for larval mortality after 24 h and 48 h, respectively. In cases where 100% mortality was observed after 24 h, the remainder of the sample, designated for 48-h assessment, was also counted. Following mortality assessment, randomly collected samples of larvae from each treatment were preserved in 80% ethanol/water and stored at 4°C prior to identification.

RESULTS AND DISCUSSION

In all cases, the black fly population was dominated by the species complex *Simulium venustum* Say/S. *verecundum* Stone and Jamnback in association with *S. tuberosum* Lundstrom, and occasionally *S. vernum* Macquart. Mortality within the control samples for each treatment was low, ranging from 0 to 4.6% over the 24-h period and 0–20% over the 48-h period. Mean larval mortality results, corrected for control mortality using Abbott's formula, are given in Tables 2 and 3. With the exception of the second 24AS treatment, nearly 100% control of black fly larvae was achieved after 24 h at the lowest treatment concentrations of 0.5 ppm (24AS) and 1.0 ppm (12AS). In keeping with the findings of other workers (Undeen and Lacey 1982), the distance over which control was achieved appeared to be related to stream size. In the stream sections treated with the 12AS formulation, control after 24 h was 100% effective up to 3,000 m, 400 m and 200 m in streams with discharge rates of 108.6 m³/min, 18.6 m³/min and 0.84 m³/min, respectively. With the 24AS formulation, the same effective distances were observed in streams with discharge rates of 37.2 m³/min,

Table 2. Mean corrected mortality (%) of Simuliidae following treatment with Vectobac-12AS.

Distance from first application point (m)	Treatment 1		Treatment 3		Treatment 5	
	Mortality (sample size)		Mortality (sample size)		Mortality (sample size)	
	24 h	48 h	24 h	48 h	24 h	48 h
10	99.2 (126)	96.5 (95)	98.8 (80)		100.0 (23)	
20	99.8 (413)	100.0 (279)	100.0 (68)		100.0 (25)	
30	98.1 (160)	100.0 (191)	100.0 (139)		100.0 (10)	
40	99.3 (143)	100.0 (180)	100.0 (197)		100.0 (10)	
50	100.0 (238)		100.0 (74)		100.0 (11)	
60	100.0 (216)		100.0 (155)		100.0 (6)	
100	100.0 (215)		100.0 (342)		100.0 (7)	
200	100.0 (198)		100.0 (98)		100.0 (40)	
320	100.0 (67)					
400			4.4 (45)	49.9 (28)	100.0 (150)	
520	100.0 (317)					
600					56.3 (151)	82.7 (151)*
700	100.0 (209)		8.9 (23)	47.7 (33)		
800					0.0 (796)	0.0 (796)*
1,000	100.0 (52)		2.9 (70)	14.3 (112)	0.0 (21)	0.0 (21)*
1,400			0.0 (63)	0.0 (35)	0.0 (138)	3.7 (138)*
2,000	99.7 (337)	100.0 (296)				
2,500			0.0 (58)	0.0 (43)		
3,000	99.2 (384)	100.0 (300)				
4,000	2.0 (356)	1.3 (154)				

* Same sample used for 24 h and 48 h assessment.

Table 3. Mean corrected mortality (%) of Simuliidae following treatment with Vectobac-24AS.

Distance from first application point (m)	Treatment 2		Treatment 4		Treatment 6	
	Mortality (sample size)		Mortality (sample size)		Mortality (sample size)	
	24 h	48 h	24 h	48 h	24 h	48 h
10	100.0 (60)		54.5 (22)	50.0 (8)	98.2 (57)	89.9 (135)
20	100.0 (57)		75.0 (8)	100.0 (23)	100.0 (98)	
30	100.0 (63)		100.0 (20)		100.0 (13)	
40	100.0 (97)		100.0 (56)		100.0 (185)	
50	100.0 (44)		100.0 (12)		100.0 (46)	
60	100.0 (34)		100.0 (69)		100.0 (22)	
100	100.0 (212)		100.0 (18)			
120					100.0 (31)	
200	100.0 (51)		100.0 (38)		100.0 (206)	
400	100.0 (23)		0.0 (7)	22.7 (22)	100.0 (30)	
600			0.0 (10)	0.0 (21)	72.3 (53)	89.5 (59)
700	100.0 (83)					
800			20.0 (10)	9.1 (11)	0.0 (62)	0.0 (80)
1,000	100.0 (210)		0.0 (30)	0.0 (9)	0.0 (94)	38.5 (180)
1,200			0.0 (25)	0.0 (22)		
1,400						
2,000	100.0 (103)					
2,500						
3,000	2.2 (125)	11.4 (22)				
4,000	0.0 (4)	0.0 (4)				

13.8 m³/min and 1.26 m³/min, respectively. Control beyond 3,000 m, 400 m and 200 m was better after 48 h, but in no case did it reach 100%.

The results observed compare favorably with those from studies using other formulations in streams with comparable discharge rates (Lacey and Undeen 1984, Lacey and Heitzman 1985). In a Newfoundland stream with a discharge rate

of 39.9 m³/min, Colbo and O'Brien (1984) observed 97–100% larval mortality at a distance of 750 m following a 1-min treatment of Teknar (600 ITU/mg) at 10 ppm. This was despite a very low water temperature of < 1°C. At 17°C similar treatment of a stream with a discharge rate of 9.5 m³/min resulted in > 95% mortality up to 200 m but only 65–70% at 350 m. Com-

parable results were observed in the present study with Vectobac-12AS and Vectobac-24AS in streams with discharge rates of less than 1.3 m³/min. Back et al. (1985) investigated the effects of a Teknar water dispersible concentrate (600 ITU) applied to a stream with a discharge rate of 114 m³/min. Despite a water temperature of 12°C, larval mortality following the application (5.86 ppm for a period of 15 min) did not reach 100% even at the 100-m sampling distance. At a discharge rate of 108.6 m³/min, the Vectobac-12AS treatment in the present study resulted in 100% mortality between 3 to 4 km downstream of the application point.

From the results of this study, it can be concluded that both Vectobac-12AS and Vectobac-24AS represent effective formulations of *B.t.i.* Although differences in application procedures, dosage rate, formulation potency, stream discharge, water temperature and target species make it difficult to make close comparisons between the results of this and other studies, both formulations exhibited improved effective carry and excellent control of black fly larvae.

ACKNOWLEDGMENTS

This study was funded by Abbott Laboratories, Chicago, Illinois. We would like to thank M. H. Colbo, Department of Biology, Memorial University of Newfoundland for his assistance in the identification of black fly larvae.

REFERENCES CITED

- Back, C., J. Boisvert, J. O. Lacoursiere and G. Charpentier. 1985. High dosage treatment of a Quebec stream with *Bacillus thuringiensis* serovar. *israelensis*: efficacy against black fly larvae (Diptera: Simuliidae) and impact on non-target insects. *Can. Entomol.* 117:1523-1534.
- Colbo, M. H. and H. O'Brien. 1984. A pilot black fly (Diptera: Simuliidae) control program using *Bacillus thuringiensis* var. *israelensis* in Newfoundland. *Can. Entomol.* 116:1085-1096.
- Depner, K. R., W. A. Charnetski and W. O. Haufe. 1980. Population reduction of the black fly *Simulium arcticum* at breeding sites in the Athabasca River, pp. 21-39. In: W. O. Haufe and G. C. R. Croome (eds.), Control of black flies in the Athabasca River. Alberta Environment, Pollution Control Division, Tech. Rep., Edmonton, Alberta.
- Gaugler, R. and J. Finney. 1982. A review of *Bacillus thuringiensis* var. *israelensis* (serotype 14) as a biological control agent of black flies (Simuliidae). *Misc. Publ. Entomol. Soc. Am.* 12(4):1-17.
- Horosko, S. and R. Noblet. 1983. Efficacy of *Bacillus thuringiensis* var. *israelensis* for control of black fly larvae in South Carolina. *J. Georgia Entomol. Soc.* 18:531-537.
- Lacey, L. A., H. Eschaffre, B. Philippon, A. Seketeli and P. Guillet. 1982. Large river treatment with *Bacillus thuringiensis* (H-14) for the control of *Simulium damnosum* s.l. in the Onchocerciasis Control Programme. *Tropenmed. Parasitol.* 33:97-101.
- Lacey, L. A. and C. M. Heitzman. 1985. Efficacy of flowable concentrate formulations of *Bacillus thuringiensis* var. *israelensis* against black flies (Diptera: Simuliidae). *J. Am. Mosq. Control Assoc.* 1:493-497.
- Lacey, L. A. and A. H. Undeen. 1984. Effect of formulation, concentration, and application time of the efficacy of *Bacillus thuringiensis* (H-14) against black fly (Diptera: Simuliidae) larvae under natural conditions. *J. Econ. Entomol.* 77:412-418.
- Lacey, L. A. and A. H. Undeen. 1986. Microbial control of black flies and mosquitoes. *Annu. Rev. Entomol.* 31:265-296.
- Margalit, J. and D. Dean. 1985. The story of *Bacillus thuringiensis* var. *israelensis* (*B.t.i.*). *J. Am. Mosq. Control Assoc.* 1:1-7.
- Molloy, D., S. P. Wraight, B. Kaplan, J. Gerardi and P. Peterson. 1984. Laboratory evaluation of commercial formulations of *Bacillus thuringiensis* var. *israelensis* against mosquito and black fly larvae. *J. Agric. Entomol.* 1:161-168.
- Pistrang, L. A. and J. F. Burger. 1984. Effect of *Bacillus thuringiensis* var. *israelensis* on a genetically defined population of black flies (Diptera: Simuliidae) and associated insects in a montane New Hampshire stream. *Can. Entomol.* 116:975-981.
- Undeen, A. H. and L. A. Lacey. 1982. Field procedures for the evaluation of *Bacillus thuringiensis* var. *israelensis* (serotype 14) against black flies (Simuliidae) and nontarget organisms in streams. *Misc. Publ. Entomol. Soc. Am.* 12(4):25-30.