Formosan Entomologist
Journal Homepage: entsocjournal.yabee.com.tw

【Research report】

兩種蘇力菌以色列變種 (Bacillus thuringiensis var. israelensis) 劑型對蚊幼蟲之殺蟲效力【研究報告】

黃榮南、羅怡珮、何兆美、黃基森、徐爾烈

*通訊作者E-mail :

Received: Accepted: 1993/03/26 Available online: 1993/06/01

Abstract

摘要

蘇力菌以色列變種 (Bacillus thuringiensis var. israelensis)是芽孢桿菌科 (Bacillacea) 、桿菌屬 (Bacilli) 中的桿狀細菌，血清型H14的以色列變種簡稱B. T. I.。本試驗以兩種不同劑型BactimosR (6000 ITU / mg 可濕性粉劑)，及VectobacR (1200 ITU / mg，粒劑及水懸浮劑)，經蒸餾水稀釋 (W / V)後，分別測定對宜蘭東鄉斑蚊 (Aedes togoi; I-Lan strain)，東鄉斑蚊 (Aedes togoi; Yang-Ming Lab. Strain)，埃及斑蚊利物浦品系 (Aedes aegypti; Liverpool strain)，埃及斑蚊台南品系 (Aedes aegypti; Tainan strain)，白線斑蚊 (Aedes aldopictus)，熱帶家蚊 (Culex quinquefasciatus; Taipei & Laboratory strain)，三斑家蚊 (Culex tritaeniorhynchus ;三齡及四齡)。蛛形翠蚊 (Tripteroides aranoides)，白腹叢蚊 (Armigeres subalbatus)，中華瘧蚊 (Anopheles sinensis)幼蟲24小時的藥效。由24小時的孳效定結果，可發現採自野外的品系較室內累代飼養品系感，而以白腹叢蚊感藥性最低；在10 ppm 下，24小時內的死亡率不及50%，另外熱帶家蚊以0.01 ppm 、0.05 ppm、0.5 ppm及1.0 ppm濃度處理的LT50 各為27.25小時、7.15小時、1.67小時及0.54小時。B.t.i. 對熱帶家蚊有迅速的毒殺效果。

Key words:
關鍵詞: 蘇力菌以色列變種、(Bacillus thuringiensis var. israelensis)、,BactimosR, VectobacR。

Full Text: PDF(0.45 MB)

中華昆蟲 13: 177-185(1993)
Chinese J. Entomol. 13: 177-185(1993)

Effectiveness of Two Formulations of *Bacillus thuringiensis* var. *israelensis* on Mosquito Larvae

Rong-Nan Huang, I-Pai Lo Department of Plant Pathology and Entomology, National Taiwan University
Chaumei Ho Department of Parasitology, National Yang-Ming Medical College
Ji-Sen Huang Bureau of Environmental Sanitation and Toxic Substance Control, Environmental Protection Administration, R.O.C.
Err-Lieh Hsu Department of Plant Pathology and Entomology, National Taiwan University

ABSTRACT

Two *Bacillus thuringiensis* var. *israelensis* formulations, Vectobac^R (Aqueous suspension & granular, 1200 ITU / mg) and Bactimos^R (Wettable powder, 6000 ITU / mg) were employed in this study for the evaluation of the efficacy of *B.t.i.* and its residual activity on the integrated pest management of mosquito larvae. Mosquito strains collected from field were revealed by the results to be more susceptible than the laboratory strain. The LC_{50} in *Armigeres subalbatus* was even over 10 ppm and its mortality was below 50% within 24 hr. Additionally, the *B.t.i.* formulations tested were exhibited by the results to be most effective against *Culex quinquefasciatus*. The LT_{50} values were 27.25 hr, 7.15 hr, 1.67 hr and 0.54 hr when dosed with 0.01 ppm, 0.05 ppm, 0.5 ppm, 1.0 ppm, respectively.

Key word: *Bacillus thuringiensis* var. *israelensis*, Vectobac^R, Bactimos^R.

兩種蘇力菌以色列變種(*Bacillus thuringiensis* var. *israelensis*)劑型對蚊幼蟲之殺蟲效力

黃榮南、羅怡珮 國立台灣大學 植物病蟲害學系 台北市羅斯福路四段113巷27號

何兆美 國立陽明醫學院 寄生蟲學科 台北市北投區立農街二段155號
黃基森 行政院環保署環境衛生及毒物管理處 台北市中華路一段41號
徐爾烈 國立台灣大學 植物病蟲害學系 台北市羅斯福路四段113巷27號

摘 要

蘇力菌以色列變種(*Bacillus thuringiensis* var. *israelensis*)是芽孢桿菌科(Bacillacea)、桿菌屬(Bacilli)中的桿狀細菌，血清型H14的以色列變種簡稱B. T. I.。本試驗以兩種不同劑型 Bactimos^{R} (6000 ITU / mg，可濕性粉劑)，及 Vectobac^{R} (1200 ITU / mg，粒劑及水懸浮劑)，經蒸餾水稀釋(W / V)後，分別測定對宜蘭東鄉斑蚊(*Aedes togoi*; I-Lan strain)，東鄉斑蚊(*Aedes togoi*; Yang-Ming Lab. strain)，埃及斑蚊利物浦品系(*Aedes aegypti*; Liverpool strain)，埃及斑蚊台南品系(*Aedes aegypti*; Tainan strain)，白線斑蚊(*Aedes albopictus*)，熱帶家蚊(*Culex quinquefasciatus*; Taipei & Laboratory strain)，三斑家蚊(*Culex tritaeniorhynchus*；三齡及四齡)，蛛形翠蚊(*Tripteroides aranoides*)，白腹叢蚊(*Armigeres subalbatus*)，中華瘴蚊(*Anopheles sinensis*)幼蟲24小時的藥效。由24小時的藥效測定結果，可發現採自野外的品系較室內累代飼養品系敏感，而以白腹叢蚊的感藥性最低；在10ppm下，24小時內的死亡率不及50%，另外熱帶家蚊以0.01 ppm、0.05 ppm、0.5 ppm及1.0 ppm濃度處理的 LT_{50} 各為27.25小時、7.15小時、1.67小時及0.54小時。*B.t.i.* 對熱帶家蚊有迅速的毒殺效果。

關鍵詞：蘇力菌以色列變種(*Bacillus thuringiensis* var. *israelensis*), Bactimos^{R}, Vectobac^{R}。

Introduction

The *Bacillus thuringiensis* var. *israelensis* (*B.t.i.*) has received extensively studies with its larvicidity against different varieties of mosquito larvae (Garcia and Desrochers, 1979; Kramer *et al.*, 1988; Molloy, 1992; Van Essen and Hembree, 1980) since it was isolated initially in Israel in 1977 (Goldberg and Margalit, 1977). Owing to the verification of environmental safety to non-target organisms and efficacy (Gharib and Hilsenhoff, 1988; Molloy, 1992; Purcell, 1981; Rishikesh *et al.*, 1983), the *B.t.i.* has also become the most widely utilized microbial agent on the control of mosquito larvae. Because of the deficiency of environmental persistence and limited larvicidal spectrum, a lot of commercial groups also acted on the formulations and genetic constructions of a new *B. thuringiensis* strain

with broadly larvicidal spectrum and a longer environmental stability (Andrews *et al.*, 1987). The utilization of microbial agents has become an alternative pest control strategy owing to the awareness of environmental contamination and resistance raised in pest by severe use of organic pesticides, and also due to its advantage of selectivity, efficacy, and biodegradability. This is specially true in the integrated pest management of public health vector of mosquito control (Garcia and Sweeney, 1986; Laird and Miles, 1985).

A number of microbial agents and commercial formulations have been evaluated during the past ten years for the potential application in mosquito control under laboratory and field condition (Lacey *et al.*, 1988a; Lacey *et al.*, 1988b; Meisch *et al.*, 1990). Among these, *B.t.i.* has been considered as the most potential candidate for the control of mosquito larvae. In practice, the larvicidal activity of *B.t.i.* was observed to be significantly affected by environmental factors, including pH, temperature, UV-light, water quality, habitats, predators and population density (Garcia and DesRochers, 1979; Ignoffo *et al.*, 1983; Lacoursiére, 1988; Molloy, 1981). Short term persistence in environmental for the control of mosquito larvae is the chief limitation of *B.t.i.* (Garcia and Sweeney, 1986; Mulla, 1985). Additionally, the efficacy of *B.t.i.* is progressively influenced by the soil constituent in the field condition as a result of absorptive interaction among a variety of particulate types and bacteria (Marshall, 1980; Ramoska *et al.*, 1982). Hence, the purpose of this study has primarily focused on the laboratory evaluation of the efficacy and residual activity of two formulations of *B.t.i.* under a simulated condition using fine sand as substrate.

Materials and Methods

Two formulations, Vectobac^{R} (Aqueous suspension & granular, Abbott Laboratories, 1200 ITU / mg) and Bactimos^{R} (Wettable powder, Biochem Chemicals, 6000 ITU / mg) were employed in this study for the evaluation of the efficacy of *B.t.i.* and its residual activity on mosquito larvae.

The field-collected larvae of *Aedes aegypti* from Tung-Kang, *Aedes albopictus* and *Culex quinquefasciatus* from Taipei were continuously cultured in the laboratory for several generations. *Culex tritaeniorhynchus* was provided by the Department of Parasitology, Yang-Ming Medical College. Mosquito larvae tested in this experiment were hatched from eggs laid by female mosquitos described as above in the laboratory. The larvae were reared in de-chlorination water with yeast in a 29.5cm × 23cm plastic disc and maintained in an incubator with the temperature 25°C. The surface scum was daily skimmed with toilet tissue and followed by a fresh addition of yeast. Adults of mosquito were blood on ICR mice. Synchronous growth of 3rd and 4th instar mosquito larvae were chosen for the following experiments. Other species of mosquito collected from field were also applied in this study for comparing their susceptibility to *B.t.i.* with that of laboratory strains.

Preliminary experiments were performed in petri dishes [10 cm dia × 5 cm h] for determination the efficacy of *B.t.i.* against mosquito larvae in various instars. Ten ml of serial dilution of *B.t.i.* in distilled water and 20 tested mosquito larvae [3rd and 4th instar] were introduced into each petri dish. Three replicates were taken for each concentrations of *B.t.i.* and the larval mortality was counted after 24 hr.

The residual activity was conducted in fine sand fundamentally according to the investigation of Ramoska *et al.* (1982) in which sand had the least effect in decreasing *B. thuringiensis* var. *israele-*

nsis efficacy against both *Ae. Aegypti* and *Cx. quinquefasciatus*. The residual activity of different formulations of *B.t.i.* was explored using Vectobac^{R}-G and Vectobac^{R}-12 AS in stainless containers. Five hundred ml of distilled water and 100g of fine sand were bounded in separated stainless container with serial 100 mg, 300 mg, 500 mg, 700 mg, 1000 mg concentrations of Vectobac^{R}-G, or 1 ml, 5 ml, and 10 ml of 100X dilute of Vectobac^{R}-12 AS. The dosages for testing Bactimos^{R} residual activity were 0.1, 0.2, 0.6, 0.8, 0.95 ppm respectively. Fifty tested larvae were then introduced into each container at day 0 and each day of subsequent observations. Dead larvae were recorded at a 12 hr interval once the tested larvae had been introduced, and the LC_{50} and LC_{95} was calculated with probit analysis. Three separate experiments were conducted for each formulation.

Results and Discussion

The comparison of the susceptibility between field-collected and laboratory strain of mosquito larvae indicated that *B.t.i.* expressed broad larvicidal spectrum and high efficiency for an extensive variety of mosquito larvae (Table 1). The *B.t.i.* formulation of Bactimos^{R} displayed high toxicity in the most concerned vectors on field collected colonies of *Ae. aegypti* (Tainan strain) and *Cx. quinquefasciatus* (Taipei strain) with the LC_{50} of 0.0032 and 0.0169 ppm, respectively; whereas *Ae. albopictus* (Taipei strain) displayed low susceptibility to this formulation with the LC_{50} of 0.2042 ppm. Unique results were obtained from *Cx. quinquefasciatus* (Laboratory strain), with the LC_{50} of 0.2743 ppm, being around 16 times less susceptible than that of

Table 1. Susceptibility of field collected and laboratory strains of mosquito larvae to *B.t.i.* (Bactimos^{R})

Species	LC_{50}(ppm)	LC_{95}(ppm)
Aedes togoi (Ilan strain)	0.0837	0.7376
Ae. togoi (Yang-Ming lab. strain)	0.1199	0.4225
Ae. aegypti (Liverpool strain)	0.0689	0.1708
Ae. aegypti (Tainan strain)	0.0332	0.2562
Ae. albopictus (Taipei strain)	0.2042	0.7521
Culex quinquefasciatus (Taipei strain)	0.0169	0.1585
Cx. quinquefasciatus (Laboratory strain)	0.2743	2.1365
Cx. tritaeniorhynchus (4th instar)	0.0238	0.0586
Cx. tritaeniorhynchus (3th instar)	0.0130	0.1737
Tripteroides aranoides	0.5494	1.0965
Armigeres subalbatus	>10	
Anopheles sinensis	0.1288	0.4107

Taipei strain (with the LC_{50} of 0.0169 ppm). It deserves further studying and keeping surveillance on its development. *Armigeres subalbatus* exhibited a high tolerance to Bactimos^R with LC_{50} higher than 10 ppm. However, the field-collected strains were indicated from this test to be more susceptible to the *B.t.i.* formulation of Bactimos^R than the laboratory strain.

Results of the efficacy of *B.t.i.* for various instars of mosquito larvae are listed in Table 2. The LC_{50} values of *Cx. quinquefasciatus* to Bactimos^R were 0.1447 and 0.3854 ppm in the early and late 4th instar larvae respectively which was apparently higher than those of *Cx. tritaeniorhynchus* to Bactimos^R, i.e. 0.0255 and 0.0091 ppm in early 4th and 3rd instar larvae respectively. The susceptibility of mosquito larvae to *B.t.i.* was generally decreased with the development of mosquito larvae. Even in the same instar, the LC_{50} of the late 4th instar was almost tripled compared to early 4th instar in this test in *Cx. quinquefasciatus*. The *B.t.i.* was almost inactive following the pupation, *e.g.* the LC_{95} was over 1000 ppm in the pupa of *Cx. quinquefasciatus*. The explanation provided by Van Essen (1980) was that due to the cessation of feeding during pupation stage, therefore reducing the possibility of *B.t.i.* consumption.

The residual activity of Vectobac^R-G and Vectobac^R-12AS on the control of mosquito are displayed in Table 3 and 4. Almost all of the mosquito larvae tested experienced 100% mortality if the dosage reached 100 mg or 1 mg per treatment in Vectovac^{R}G and Vectobac^R-12 AS respectively; However, the larval mortalities of Vectobac^R-12AS to *Ae. albopictus* were only 82%, 87% and 99% following 12, 24, or 36hr of exposure respectively.Apparently, Vectobac^R-G was more effective than the Vectobac^R-12AS in killing mosquito larvae, and its residual activity could persist over 1-3 weeks (Table 3 & 4). In contrast, only 87% mortality was observed in 24 hr exposure of *Ae. albopictus* to Vectobac^R-12AS and the residual activity dramatically declined after 1 week (Table 3). In Bactimos^R, the residual activity was not as effective as the Vectobac^R. The Bactimos^R is indicated from table 5 to be most effective against *Ae. aegypti* in which mortality in 12 hr was more than 90% when dosed with 0.2 ppm. However, the mortality dosed with higher as 0.6 ppm were even lower than 50% within 12 hr in *Cx. quinquefasciatus* and *Ae. albopictus*. The residual activity of Bactimos^R was lowered than 10% in day 7 for all of the tested larvae. The mechanism involving the reduction of larvicidal effectiveness of *B.t.i.* by soil constituents has not been demonstrated; however, several possibilities could be considered; first, chemical deterioration of the toxin of *B.t.i.*; second, physical adherence and sedimentation of toxin-containing sporangia with soil particles;

Table 2. Effecicacy of *B. thuringiensis* var *iseraelensis* (Bactimos^R) on different instar larvae of *Cx. quinquefasciatus* and *Cx. tritaeniorhynchus*

Species	Stage	LC_{50}(ppm)	LC_{95}(ppm)
Cx. quinquefasciatus			
	early 4th instar	0.1447	0.6679
	late 4th instar	0.3854	2.0893
	pupa		>1000
Cx. tritaeniorhynchus			
	early 3rd instar	0.0091	0.0793
	early 4th instar	0.0255	0.0714

Table 3. Residual activity of *B.t.i.* Vectobac^{R}-G on the control of mosquito larvae

Dosage ppm	Time[1)]	Mortality(%) in different observation time				
		12hr	24hr	36hr	48hr	60hr
Cx. quinquefasciatus						
1000	Day 1	100	100	100	100	—
500	Day 1	100	100	100	100	—
300	Day 1	100	100	100	100	—
100	Day 1	100	100	100	100	—
	Day 6	91	97	99	100	—
	Day 62	1	5	6	—	—
Ae. albopictus						
500	Day 1	100	100	100	—	—
300	Day 1	100	100	100	—	—
100	Day 1	100	100	100	—	—
	Day 11	45	47	58	—	—
Ae. aegypti						
500	Day 1	100	100	100	100	100
300	Day 1	100	100	100	100	100
100	Day 1	100	100	100	100	100
	Day 9	99	99	99	100	100
	Day 15	100	100	100	100	100
	Day 21	91	98	99	99	100
	Day 40	31	66	76	—	—

1) : indicates the tested larval was introduced into each petri dish on the day following applicaton of the *B.t.i.*

and third, interference with feeding behavior (Van Essen and Hembree, 1982). Results from this study tended to support the second mechanism, since no effect was observed in the control experiment in which the same substrate with no *B.t.i.* added was employed.

The efficacy of *B.t.i.* formulation of Bactimos^{R} on mosquito larvae is listed in Table 6 with different exposure periods. All of the tests from three various species of mosquito demonstrated that the efficacy of *B.t.i.* formulation of Bactimos^{R} to mosquito larvae could be effectively enhanced by lengthening the exposure period of mosquito larvae to *B.t.i.* The LC_{50} was more than 3 times improved once the exposure periods were doubled, *e.g.* the LC_{50} ranged from 0.0169 ppm in 24 hr exposure down to 0.0048 ppm in 48 hr exposure in *Cx. quinquefasciatus*. Similar results were observed for *Ae. albopictus*, the LC_{50} ranged 0.2042 ppm in 24 hr exposure down to 0.0623 ppm in 48 hr exposure and in *Ae. aegypti*, the LC_{50} was 0.0991 ppm in 12 hr exposure and 0.0332 ppm in 24 hr exposure. From these investigations, if time is not a limitation and based on the residual activity tested above, longer exposure period is suggested here for minimizing the concentration required for killing mosquito larvae in view of environmental contamination and economical feasibility. Owing to the short residual effect of *B.t.i.* being the primary weak point, the critical efforts in the future should be made toward obtaining a slow-released formulation of *B.t.i.* by physical, chemical or genetical means.

Acknowledgments

The authors are grateful to the Environmental Protection Administra-

Table 4. Residual activity of *B.t.i.* Vectobac®-12AS on the control of mosquito larvae

Dosage	Time[1)]	Mortality(%) in different observation time					
ppm		12hr	24hr	36hr	48hr	60hr	72hr
Cx. quinquefasciatus							
10	Day 1	100	100	100	100	100	100
7	Day 1	100	100	100	100	100	100
5	Day 1	100	100	100	100	100	100
3	Day 1	100	100	100	100	100	100
1	Day 1	100	100	100	100	100	100
	Day 15	0	29	49	63	77	81
	Day 62	0	1	1	—	—	—
Ae. albopictus							
5	Day 1	100	100	100	—	—	—
3	Day 1	100	100	100	—	—	—
1	Day 1	82	87	99	—	—	—
Ae. aegypti							
5	Day 1	100	100	100	100	100	—
3	Day 1	100	100	100	100	100	—
1	Day 1	100	100	100	100	100	—
	Day 7	100	100	100	100	100	—
	Day 13	77	85	92	92	92	—
	Day 32	5	19	26	—	—	—

1) : indicates the test larval was introduced into each petri dish on the day following application of the *B.t.i.*

Table 5. Residual activity of *B.t.i.* Bactimos® on the control of mosquito larvae

Dosage	Time[1)]	Mortality(%) in different observation time					
ppm		12hr	24hr	36hr	48hr	60hr	72hr
Cx. quinquefasciatus							
0.10	Day 1	13.3	30.7	44.2	48.1	80.2	80.2
0.20	Day 1	36.7	83.3	88.3	91.7	90.2	90.2
0.60	Day 1	20.0	86.7	98.3	98.3	96.2	98.0
0.80	Day 1	61.7	65.9	80.8	91.4	95.7	95.7
0.95	Day 1	72.3	82.9	89.4	93.6	93.6	98.2
	Day 7	0.0	0.0	0.0	2.3	2.3	2.3
Ae. albopictus							
0.10	Day 1	0.0	21.2	72.7	72.7	82.2	92.0
0.20	Day 1	6.8	50.0	97.7	97.7	98.2	98.2
0.60	Day 1	50.0	91.3	100.0	—	—	—
	Day 7	0.0	0.0	0.0	1.5	1.5	2.1
Ae. aegypti							
0.10	Day 1	52.1	69.5	78.3	78.2	86.9	100.0
0.20	Day 1	97.1	97.1	97.1	97.1	100.0	—
0.60	Day 1	97.1	97.1	97.1	97.1	100.0	—
	Day 7	0	5	5.2	5.2	12.3	12.3

1) : indicates the test larval was introduced into each petri dish on the day after the *B.t.i.* was applied

Table 6. Effecicacy of *B.t.i.* (Bactimos®) on the control of mosquito larvae of *Cx. quinquefasciatus* and *Ae. albopictus, Ae. aeqypti* with various observation times

Species	Observation time (hr)	LC_{50}* (ppm)	LC_{95}* (ppm)
Cx. quinquefasciatus	1	0.7288	7.0843
	12	0.0196	0.3189
	24	0.0169	0.2083
	36	0.0144	0.1758
	48	0.0048	0.1122
Ae. albopictus	24	0.2042	0.7521
	48	0.0623	0.1844
Ae. aegypti	12	0.0991	0.1825
	19	0.0523	0.2745
	24	0.0332	0.2562

tion, R. O. C., for a grant in support of this work.

References

Andrews, Jr. R. E., R. M. Faust, H. Wabiko, K. C. Raymond, and L. A. Bulla, Jr. 1987. The biotechnology of *Bacillus thuringiensis*. CRC Critical Reviews in Biotechnology. 6: 163.

Garcia, R., and A. W. Sweeney. 1986. The use of microbial pathogens for the control of mosquitoes. Agric. Ecosystems Environ. 15: 201–208.

Garcia, R., and B. Desrochera. 1979. Toxicity of *Bacillus thuringiensis* var. *israelensis* to some California mosquitoes under different conditions. Mosq. News 39: 541–544.

Gharib, A. H., and W. L. Hilsenhoff. 1988. Efficacy of two formulations of *Bacillus thuringiensis* var. *israelensis* (H-14) against Aedes vexans and safety to non-target macroinvertebrates. J. Am. mosq. Cont. Assoc. 4: 252–255.

Goldberg, L. J., and J. Margalit. 1977. A⁻bacterial spore demonstrating rapid larvicidal activity against *Anopheles sergentii, Uranotaenia unguiculata, Culex univittatus, Aedes aegypti and Culex pipiens*. Mosq. News 37: 355–358.

Ignoffo, C. M., C. Garica, M. Kroha, and T. L. Couch. 1983. The effects of temperature and water on the insecticidal activity and spore viability of a wettable powder foundation of *Bacillus thuringiensis* var. *israelensis*. J. Kansas Entomol. Soc. 56: 88–92.

Lacey, L. A., C. M. Lacey, and L. E. Padua. 1988a. Host range and selected factors influencing the mosquito larvicidal activity of the PG-14 isolate of *Bacillus thuringiensis* var. *morrisoni*. J. Am. Mosq. Control Assoc. 4: 39–43.

Lacey. L. A., C. M. Lacey, B. Peacock, and I. Thiery. 1988b. Mosquito host range and field activity of *Bacillus sphaericus* isolate 2297 (serotype 25). J. Am. Mosq. Control Assoc. 4: 51–56.

Lacoursière J. O., and G. Charpentier. 1988. Laboratory study of the influence of water temperature, and pH on *Bacillus thuringiensis* var. *israelensis* efficacy against black fly larvae (Diptera: Simulidae). J. Am. Mosq. Control Assoc. 4: 64–72.

Marshall, K. C. 1980. Absorption of microorganisms to soils and sediments. pp. 317–320 in G. Bitton and K. C. Marshall, eds. Adsorption of microorganisms to surfaces. John Wi-

ley & Sons, New York.

Meisch, M. V., M. F. Finch, A. A. Weathersbee, J. W. Jones, D. G. Bassi, and D. E. Bowles. 1990. Efficacy of various *Bacillus thuringiensis* var. *israelensis* formulations against *Psorophora columbiae* larvae as assessed in small rice plots, 1984–88. J. Am. Mosq. Control Assoc. 6: 93–95.

Molly, D. P. 1992. Impact of the black fly (Diptera: Simuliidae) Control agent *Bacillus thuringiensis* var. *israelensis* on chironomids (Diptera: Chironomidae) and other non-target insects: result of ten field trials. J. Am. Mosq. Control Assoc. 8: 24–31.

Molly, D. P., D. R. Gaugler, and H. Jamnback. 1981. Factors influencing efficacy of *Bacillus thuringiensis* var. *israelensis* as a biological agent of black fly larvae. J. Econ. Entomol. 74: 61–64.

Mulla, M. S. 1985. Field evaluation and efficacy of bacterial agents and their formulations against mosquito larvae pp. 227–250 in M. Laird and J. M. Miles, eds. Integrated mosquito control methodologies Vol. 2. Academic Press, London.

Perich, M. J., P. M. Clair, and L. R. Boobar. 1990. Integrated use of Planaria (*Dugesia dorotocephala*) and *Bacillus thuringiensis* var. *israelensis* against *Aedes taeniorhynchus*: A Laboratory Bioassay. J. Am. Mosq. Control Assoc. 6: 667–671.

Purcell, B. H. 1981. Effects of *Bacillus thuringiensis* var. *israelensis* on *Aedes taeniorhynchis* and some non-target organisms in the salt marsh. Mosq. News 41: 476–512.

Ramoska, W. A., S. Watts, and R. E. Rodriguez. 1982. Influence of suspended particulates on the activity of *Bacillus thuringiensis* serotype H-14 against mosquito larvae. J. Econ. Entomol. 75: 1–4.

Rishikesh, N., H. D. Burges, and M. Vandekar. 1983. Operational use of *Bacillus thuringiensis* serotype H-14 and environmental safety. WHO/VBC/83.871.

Van Essen, F. W., and S. G. Hembree. 1980. Laboratory bioassay of *Bacillus thuringiensis* var. *israelensis* against all instars of *Aedes aegypti* and *Aedes taeniorhynchus* larvae. Mosq. News 40: 424–431.

Van Essen, F. W., and S. G. Hembree. 1982. Simulated field studies with four formulations of *Bacillus thuringiensis* var. *israelensis* against mosquites: residual activity and effect of soil constituents. Mosq. News 42: 66–72.

收件日期：1992 年 11 月 24 日
接受日期：1993 年 3 月 26 日