EVALUATION OF SLOW-RELEASE FORMULATIONS OF TEMEPHOS (ABATE®) AND *BACILLUS THURINGIENSIS* VAR. *ISRAELENSIS* FOR THE CONTROL OF *AEDES AEGYPTI* IN PUERTO RICO

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ABSTRACT. Formulations of temephos (Abate®) and *Bacillus thuringiensis* var. *israelensis (B.t.i.)* on corncob and dried coconut husk carriers were tested for slow-release insecticide properties against *Aedes aegypti* larvae in Puerto Rico. Granular formulations of 5% and 10% temephos gave continuous larval control in used automobile tires for 27 to 124 days and 34 to 162 days, respectively. Temephos on radial sections of corncob and coconut husk chips gave good larval control in tires for 27 to 63 and 61 to 134 days, respectively, depending on the size of the carrier. Small (2–3 g) and large (3–5 g) coconut husk chips tested in 167 liter drums provided continuous control for 55 to 105 days, respectively. Granular formulations of *B.t.i.* controlled *Ae. aegypti* in tires for 19 to 33 days, and *B.t.i.* briquets exhibited larvicidal activity in large containers for 26 to 78 days.

INTRODUCTION

In Puerto Rico, as in other areas of Latin America and the Caribbean, many of the most important habitats of *Aedes aegypti* (Linn.) larvae are difficult to control. Collection and storage of rainwater is common. Storage containers such as water storage drums, tubs, buckets and cisterns are major sources for this vector mosquito (Moore et al. 1978, Nathan and Giglioli 1982, Giglioli 1979, Chadee 1984). Other larval habitats that are common and produce large populations of *Ae. aegypti* in Puerto Rico are used-tire yards, automobile wrecking yards, junk accumulations in vacant lots and roadsides, and cemetery vases and urns (San Juan Laboratories, unpublished data). Environmental sanitation is often not feasible because of utilitarian, logistic or economic reasons.

Because *Ae. aegypti* are domestic mosquitoes, many toxicants, for human safety reasons, cannot be used for their control. Studies by Gaines (1969), Laws et al. (1967, 1968), however, have shown that temephos (Abate®) can be used safely in domestic situations for controlling this species.

Recent studies in Illinois have shown that temephos can be formulated onto selected size-grades of corncob granules to achieve specific rates of release. These granular formulations were effective in controlling *Aedes vexans* (Meigen) in a variety of sites ranging from river floodplains to upland cattail sloughs and right-of-way ditches (Dr. Daniel Brown, Macon County Mosquito Abatement District, Decatur, IL, personal communication). Properties of the granular corncob carrier allowed for maximum versatility regarding formulation, vegetation penetration and adaptability to various aerial and ground dispersal equipment.

The purpose of this study was to test the duration of effective *Ae. aegypti* larval control using a slow-release formulation of temephos on corncob carriers. Since corncobs are not readily available in Puerto Rico or other tropical areas, coconut husks were selected as a substitute. Because the continued use of one insecticide increases the chances of resistance, a slow-release formulation of *Bacillus thuringiensis* var. *israelensis (B.t.i.)* was tested.

MATERIAL AND METHODS

The temephos formulations used were prepared by Mosquito Management Consultants, Decatur, IL[3] (Fig. 1). Briefly, the desired active ingredient concentration of temephos was made using a mineral oil base. This mixture was then topically applied to the carriers by means of two spray nozzles fitted on a grain auger conveyer system. Corncob granules in the range of 4- to 8-mesh (U.S. Standard) and weighing 0.05 to 0.5 g, were formulated to give an active ingredient concentration of temephos of 0.016 mg per granule for the 5% formulation and 0.03 mg per granule for the 10% formulation. Full-sized corncobs and dried coconut husks that were cut into small chips were treated with temephos to give an active ingredient concentration of 0.25 mg. The full-sized corncobs were then cut radially into 5- to 8-mm sections for future testing. The corncob sections, and the coconut chips were formulated so that a full sized piece when applied to a 208 liter drum

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[3] Use of trade names and commercial sources is for identification only and does not imply endorsement by the Public Health Service or the U. S. Department of Health and Human Services.

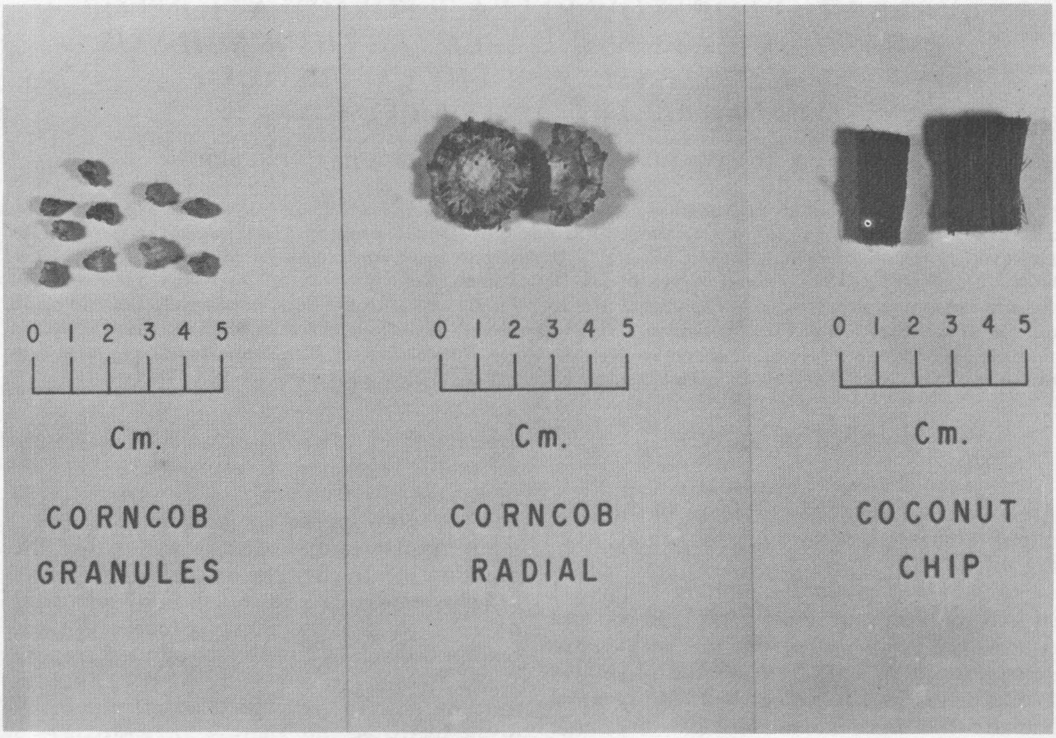

Fig. 1. Carriers used to test the duration of temephos activity for the control of *Aedes aegypti* larvae.

would give a final concentration of temephos in water less than or equal to 1 ppm as recommended (American Cyanamid, Abate Technical Bulletin, 5th edition).

The *B.t.i.* products used were supplied by Biochem Products, Montchamin, DE. Bactimos® granules are formulated on 12- to 14-mesh (U.S. Standard) corncob to give a 2.5% active ingredient concentration of *B.t.i.* The Bactimos® briquets are a floating, sustained release formulation containing 5% *B.t.i.*

Corncob and coconut carriers impregnated with temephos and corncob granules and Bactimos briquet with *B.t.i.* were tested in used automobile tires, 167 liter drums, and a 757 liter tank on the grounds of San Juan Laboratories, Centers for Disease Control. The tires were placed under trees protected from direct sunlight. The drums and tank were placed in a protected area behind a laboratory building where exposure was only to morning sunlight.

The experimental design was as follows. A free breeding population of *Ae. aegypti* was established in each container and monitored for 30 days before each treatment. One container was maintained as an untreated control for each formulation or carrier tested. After the insecticide was added, all containers were visually inspected daily for live *Ae. aegypti* larvae, and on the same day each week, 100 laboratory reared third and fourth-instar *Ae. aegypti* larvae were added to each container to confirm the duration of larvicidal activity. At the same time, each week, water was added to each container to a predetermined level to maintain a standard volume and to flood any eggs deposited above the receding water line. This procedure continued until a container was positive with live larvae for 2 consecutive days after the weekly challenge with laboratory reared larvae.

RESULTS

In the first trial, 1, 3, 5, and 10 corncob granules of the 5% and 10% granular formulations of temephos were tested per tire. The 5% formulation gave continuous control for 27, 51, 87 and 124 days with 1, 3, 5 and 10 granules, respectively (Fig. 2). Similarly, the 10% formulation provided effective larval control for 34, 78, 108 and 162 days with 1, 3, 5 and 10 granules per container, respectively.

Two replicate tests were carried out consecutively in automobile tires with temephos formulated on corncob radial sections and coconut chips (Table 1). A single ½ corncob radial section per tire (weight, 0.3–0.5 g) resulted in con-

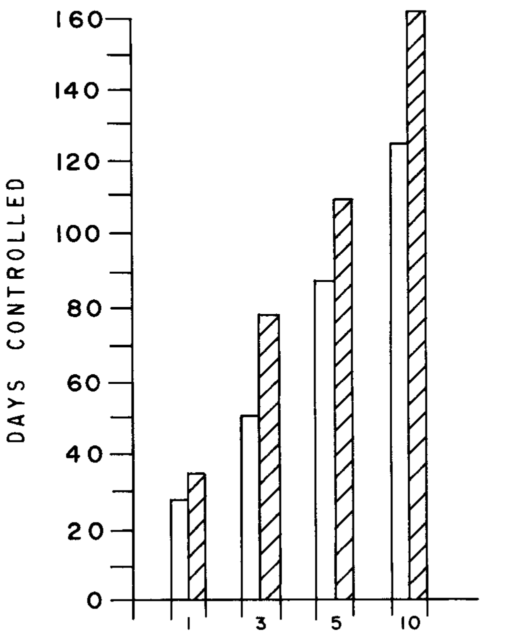

Fig. 2. Larvicidal activity of 5 and 10 percent temephos formulated on corncob granules. The open bar represents 5% temephos (0.016 mg per granule) and the slashed bar represents 10% temephos (0.03 mg per granule).

tinuous larval control for 27 and 30 days in the two trials compared to 58 and 63 days for single full radial sections (0.6–0.9 g per tire). The coconut chip carriers provided effective larval control in automobile tires twice as long as the corncob sections (Table 1). The small chip (2–3 g) killed larvae for 61 and 69 days in the two trials compared to 123 and 134 days for the large chip (3–5 g).

Only coconut chip carriers were tested in drums and a 757 liter tank (Table 2). The small single chip (2–3 g) provided larval control for 55 days in the drums compared to 105 days for a single larger chip (3–5 g). A large chip killed larvae for 44 days in the tank.

In automobile tires, 0.1 g per tire of the granular formulation of *B.t.i.* did not have larvicidal activity (Table 3). Larval control was obtained for 19 and 33 days with 0.3 g and 0.5 g per tire, respectively. The *B.t.i.* briquets gave effective larval control in individual tires for 43, 48, 69, and 75 days when ⅛, ¼, ½, and a whole briquet were used, respectively (Table 3). In the drums, ½ of a briquet provided larvicidal activity for 26 days compared to 54 days when a whole briquette was used (Table 3). Surprisingly a whole briquet killed larvae for 78 days in the 757 liter tank.

DISCUSSION

Although elimination of the larval habitat is the preferred method of controlling *Ae. aegypti* in Puerto Rico, there are many situations where this is not possible. We have shown that temephos, formulated on corncob or coconut husk carriers, can be used as a practical alternative for effective larval control.

The 5% and 10% granular formulations of temephos were tested as a prelude to field studies in sites having numerous water-holding containers such as tire yards, automobile-wrecking yards, cemeteries, roadside areas and

Table 1. Duration of *Aedes aegypti* larval control in automobile tires using temephos formulated on corncob radials and coconut husks.

	Formulation		Duration of control (days)	
Carrier	Weight of carrier	Temephos (mg)*	Trial 1	Trial 2
Corncob	½ radial (0.3–0.5 g)	0.12	27	30
	Full radial (0.6–0.9 g)	0.25	58	63
Coconut husk	Small chip (2–3 g)	0.12	61	69
	Large chip (3–5 g)	0.25	123	134

* mg/radial or/chip.

Table 2. Duration of *Aedes aegypti* larval control in large containers using temephos formulated on coconut husks.

	Formulation		
Container type	Weight of carrier	Temephos (mg)*	Duration of control (days)
167 liter (44-gal) drum	Small chip (2–3 g)	0.12	55
167 liter (44-gal) drum	Large chip (3–5 g)	0.25	105
757 liter (200-gal) tank	Large chip (3–5 g)	0.25	44

* mg/chip.

Table 3. Duration of *Aedes aegypti* larval control in automobile tires and large containers using *Bacillus thuringiensis* var. *israelensis* formulations.

Carrier	Container	Treatment rate	Days of control
Granule	Tire	0.1 g/tire	No activity
Granule	Tire	0.3 g/tire	19
Granule	Tire	0.5 g/tire	33
Briquet[1]	Tire	1/8/tire	43
Briquet	Tire	1/4/tire	48
Briquet	Tire	1/2/tire	69
Briquet	Tire	1/tire	75
Briquet	167 liter drum	1/2/drum	26
Briquet	167 liter drum	1/drum	54
Briquet	757 liter tank	1/tank	78

[1] Briquets are donut-shaped.

vacant lots. The physical attributes of the granular formulations make them well suited for use at these sites since they can be broadcast by hand or machine and can penetrate dense vegetation. Both 5% and 10% formulations gave excellent results, with larvicidal activities ranging from 28 to 162 days depending on the number of granules in each container. Ongoing studies will determine the application rate and dispersal efficiency of the corncob granule formation under natural field conditions.

Brooks et al. (1965), reported 13 weeks of larvicidal activity with temephos formulated on sand and on bentonite in storage drums in the U. S. Virgin Islands. Similarly, Chadee (1984) reported 4–6 weeks (28–42 days) control in drums containing *Ae. aegypti* in Trinidad. Temephos formulated on coconut chips in our study gave effective larval control in similar drums of 55 days (8 weeks) for small chips and 105 days (15 weeks) for larger chips. The toxicant on coconut chips gave a longer duration of control than the corncob radial sections when tested in tires. This is a most promising method since coconuts are very abundant in the tropics and cost little in most parts of the world where this vector occurs.

The cost of a control program is a major consideration in most areas where *Ae. aegypti* occurs. The low cost and simplicity of temephos formulation on corncob and coconut husk carriers, and the ease of application, make this a very economical and logistically simple way to control mosquitoes. Thus, the only equipment necessary for field application is a plastic bucket and a pair of gloves. The duration of larvicidal activity observed with theses carriers suggests that effective control can be achieved by applications of toxicant every 2 to 3 months, depending on the formulation.

In the present study, *B.t.i.* was also effective in controlling *Ae. aegypti* larvae, but the duration of control did not approach that observed with the temephos formulations. As with temephos, the efficacy of the granular formulation of *B.t.i.* in tires was affected by the amount of toxicant used. Insecticidal activity did not occur when 0.1 g of granular *B.t.i.* was added, although 100% mortality was noted after a 48-hr exposure to 0.3 g and 0.5 g per tire. Similar results on *B.t.i.* activity were reported by Van Essen and Hembree (1980). The Bactimos® briquet in large water-holding containers gave the longest duration of larvicidal activity, 43 to 75 days, again depending on the amount used. Lacey et al. (1984), found that the Bactimos® briquets controlled *Culex quinquefasciatus* Say larvae for 21 days (3 weeks).

Since many of the containers that harbor *Ae. aegypti* are in domestic environments, it is essential that insecticides used for this species have low toxicity for both humans and domestic animals. Law et al. (1967), found that human volunteers tolerated temephos doses of 256 mg/day for 5 days and 64 mg/day for 4 weeks without clinical symptoms or side effects. They also reported no elevation of red blood cell or plasma cholinesterase in these human subjects. Field studies in Puerto Rico failed to reveal any ill effects when temephos was added to stored drinking water in a community of 2000 people (Law et al. 1968). Garcia et al. (1980), have shown that non-target organisms such as aquatic insects, crustaceans, amphibians, fish and mammals are unaffected by *B.t.i.* The label on this toxicant indicates that it is also safe for use in potable water.

Our results indicate that both toxicants may be effective for long-term control of *Ae. aegypti* larval populations. In addition, their use in an integrated control program is beneficial since alternating insecticides should delay development of resistance in the mosquito populations. Long-term field studies are planned to determine feasibility under natural conditions.

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