CONTROL OF SALT-MARSH MOSQUITOES WITH ABATE[1] INSECTICIDE AT COOMBABAH LAKES, QUEENSLAND, AUSTRALIA

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ABSTRACT. 4.0 and 4.4% Abate insecticide-sand granule formulations, each applied aerially at 1 lb of formulated product/acre, were 91% and 71% effective against larval *Aedes vigilax* Skuse and *Culex sitiens* Wiedemann respectively in a 1971 trial. In 1972, even higher mortalities were indicated. At the dosages used, Abate insecticide caused minimal mortality of non-target organisms tested in natural salt-marsh conditions. Ninety-nine percent kill of larval *C. annulirostris* Skuse and 87% mortality of *C. fatigans* Wiedemann exposed in small containers were also recorded. The granules penetrated dense mangrove cover and open country with equal efficiency. A successful application was carried out during a 15–20 knot wind. The insecticide was found to have little residual life in simulated salt-marsh conditions.

Introduction. Abate insecticide, O,O, O',O'-tetramethyl O,O'-thiodi-p-phenylene phosphorothioate) has proved useful in treatment of potable water against *Aedes aegypti* L., a vector of yellow, dengue and haemorrhagic fevers (Laws *et al.*, 1968). It has proved extremely toxic to any larval mosquitoes tested (Gras and Rioux, 1969; Hooper, 1966) but not to man (Laws *et al.*, 1967) and other animals (Mulla, 1966; Gaines *et al.*, 1967).

Aedes vigilax (Skuse) is a vicious pest of man in coastal Australia, New Guinea and parts of the Oriental region and has been incriminated as a vector of Ross River virus in Australia possibly responsible for outbreaks of epidemic polyarthritis (Doherty, 1972). *Culex sitiens,* commonly found breeding in association with *Ae. vigilax* is presently notable at pest rather than at vector status. Two fresh water breeding species were also treated. *Culex annulirostris* Skuse is probably the most important arbovirus vector in Australia with no fewer than 10 different viruses isolated from it (Doherty, 1972). In Australia, *C. fatigans* Wiedemann has probably minor status as a vector of arboviruses (Carley *et al.*, 1973) but is a major pest. All species, except *C. sitiens,* are natural vectors of filariasis in parts of Australasia (Iyengar, 1965).

Abate was examined in salt-marsh country near the Gold Coast tourist area of Queensland with respect to mosquito control and environmental effects.

Materials and Methods. The study area, 12 acres of mangrove (*Avicennia marina*)—salt couch (*Sporobolus virginicus*) country bordering Lake Coombabah (Fig. 1) lies 45 miles south of Brisbane. Field trials, conducted on May 5–6, 1971 and March 5–9, 1972, were supplemented by laboratory experiments.

The granule formulation. Abate was adsorbed onto —25 +52 British Standard Sieve (600 to 300 μ) sand granules but technical problems, since rectified, resulted in understrength 4.0 and 4.4 percent formulations being produced as Abate 5G (5 percent).

Application. 'Agcat' 300 aircraft fitted with a modified Grumman aerofoil spreader, flying at 50 ft and 90 m.p.h., dispensed Abate insecticide sand granules at 1 lb of formulated product/acre over a 33-ft swath.

Field Assessment. Pre- and post-application estimations of immature mosquitoes were made using 10 scoops of a 4.25 inch diameter ladle in large (>100 ft^2; 3 to 6 inches deep) and medium (1–100 ft^2;

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Fig. 1.—Typical salt-marsh country at Coombabah.

3 to 6 inches deep) pools or by direct counting in small pools (<1 ft^2; 1 to 2 inches deep).

In 1972, heavy rain following treatment prevented post-application assessment of individual breeding pools. The pre- and post-application assessments were therefore not directly comparable so the efficacy of the insecticide was judged by comparing post-application assessments of treated and untreated populations.

Sixteen cardboard containers, each with 50 *C. annulirostris* (1971) and 10 with 50–100 *C. fatigans* (1972) in 500 ml of clean water were exposed, with similar untreated containers kept 50 yards from the treatment zone.

An untreated check area, 200 yards away from the treatment zone, was monitored in both years. All post-application assessment was carried out at 24 hours.

Local aquatic non-target organisms were exposed in 8x6x6 inch stocking mesh cages in pools in the treatment zone. Similar groups of organisms were maintained in the untreated area.

Laboratory Studies. The residual life of Abate insecticide sand granules was determined in glass crystallizing bowls containing 250 ml. of clean Brisbane tapwater, clean saltwater and saltwater with a mud substrate from Coombabah. Granules were added to approximate the rates of 0.5, 1, 2 and 5 lb product/acre for each set of conditions. A fresh batch of 20 IV instar larvae (*Aedes aegypti* in tapwater; *Aedes vigilax* in saltwater) was added to each bowl every 24 hours for daily assessment.

Standard methods comparable to those for determination of susceptibility of mosquito larvae to insecticides (W.H.O., 1963) were used to determine susceptibility of aquatic non-target organisms. Abate was diluted in acetone.

RESULTS. *Aerial application of granules.* Successful applications, judging by larval mortality and granule distribution, were made in both trials despite a 15–20 knot crosswind in 1972. Lateral displacement of granules in this latter trial was approximately three swath widths. An average of 44 granules/ft^2 was recorded in 68 sampling trays placed at random in the treatment area. There was no significant difference ($P<0.01$) between the number of granules/ft^2 falling on open country or on the ground beneath dense mangrove cover.

There were indications of a bimodal distribution within the 33-foot swath used. The total swath was 54 feet but dosage at extremities was low.

Mosquito Mortality. In 1971, percentage mortality of *Ae. vigilax* and *C. sitiens* were 91 and 71 respectively (Table 1). The results in 1972 were interpreted as indicating even higher mortality as the survivors were mainly first instar *Ae. vigilax* which probably hatched during the flooding the night after treatment. The mortalities of larval *C. annulirostris* and *C. fatigans* exposed in small containers were 99 percent and 87 percent respectively (Table 2).

In both applications, only 2 out of 66 sites were not treated satisfactorily due to either poor granule distribution or pilot error. One pool of *C. sitiens* and one container of *C. fatigans* were missed by the granules which explained the lower mortality figures. On the other hand, all of the 21 small pools (hoof prints and other small depressions) monitored in 1971 were effectively treated.

Untreated check areas in some instances were not examined quantitatively and the populations are indicated by plus signs.

Residual Life of Abate. Mortality at one day for all conditions tested (clean tapwater, clean saltwater and saltwater with mud substrate at rates approximating 0.5, 1, 2 and 5 lb formulated product/acre) was 100 percent. After 4 weeks, the granules at all rates were still killing greater than 50 percent of larvae introduced into both clean tap and saltwater. Granules added to saltwater with a mud substrate lasted approximately 48 hours at rates approximating 0.5, 1 and 2 lb formulation/acre whereas a life expectancy of one week was indicated at the rate equivalent

TABLE 1.—Mortality of salt-marsh mosquito larvae in situ 24 hours after treatment with Abate 5G insecticide at 1 lb formulation/acre.

Date	Species	Treatment (lbs active ingredient/acre)	Number of sites	Size of pool	Population	
					Pre-treatment	Post-treatment
1971	*Aedes vigilax*	0.04	21	small	270 IV instar	1 IV instar 23 pupae
	Culex sitiens	0.04	7	medium-large	249 IV instar 5 pupae	47 IV instar 27 pupae
	mixed	untreated	20	small, medium, large	+++	+++
1972[1]	*Aedes vigilax*	0.044	21	small, medium, large	56 I instar 119 II instar 218 III instar 154 IV instar 254 pupae	408 I instar 2 II instar
	Culex sitiens[2]	0.044	8	small, medium, large	23 I instar 214 II instar 24 III instar 29 IV instar 8 pupae	22 I instar 7 II instar
	mixed	untreated	20	small, medium, large	+++	192 I instar 385 II instar 21 III instar 14 IV instar 7 pupae

[1] Pre-treatment population combined counts from individual pools—post-treatment assessed after flooding of test area.

[2] Breeding in conjunction with *Ae. vigilax* in the eight sites.

to 5 lb formulation/acre. Mortality in untreated bowls was negligible.

Non-target Organisms. Field and laboratory data are presented together (Table 3). At the dosages employed, Abate insecticide showed little toxicity to groups of Crustacea, Insecta, Mollusca and to mosquito fish.

DISCUSSION. Many field evaluations of Abate have been published, especially in North America (Hagmann, 1966; Lake *et al.*, 1967; Moore and Breeland, 1967; Schober, 1967; Tawfik and Gooding, 1970; von Windeguth *et al.*, 1971). Authorities of the Illinois, Michigan and Minnesota districts (Buzicky, 1970) and the New Jersey area (Vannote, 1971) have used Abate regularly with success against mosquitoes. However, general organophosphate resistance has made this group of

TABLE 2.—Mortality of freshwater mosquito larvae exposed in 4.8 inch diameter containers 24 hours after treatment with Abate 5G insecticide at 1 lb formulation/acre.

Date	Species	Treatment (lbs a.i./acre)	Number of containers	Population	
				Pre-treatment	Post treatment
1971	*Culex annulirostris*	0.04	16	677 IV instar	8 pupae
	Culex annulirostris	untreated	5	300 IV instar	285 IV instar
1972	*Culex fatigans*	0.044	10	74 I instar 261 II instar 142 III instar 74 IV instar	21 II instar 50 IV instar
	Culex fatigans	untreated	10	+++	+++

TABLE 3.—Mortality of non-target organisms on exposure to Abate insecticide, assessed at 24 hours.

	Organism	Treatment[1]	Number of containers	Population Pre-treatment	Population Post-treatment
Insecta	Odonata nymphs (f. Libellulidae)	.04 lb a.i./ac	2	35	28
	(f. Coenagrionidae)	.04 lb a.i./ac	2	15	15
	mixed Odonata	untreated	1	20	20
Mollusca	Pulmonata (*Salinator fragilis*)	.04 lb a.i./ac	field observation	+++	+++
	Opisthobranchiata (*Ereolania* sp. (undescribed))	.001–.1 ppm	8	40	40
Pisces	*Gambusia affinis*	.04 lb a.i./ac	field observation	12	12
		.01 ppm	2	6	6
Crustacea	Ostracoda	.04 lb a.i./ac	4	12	12
	Copepoda	.04 lb a.i./ac	field observation	+++	+++
	Metapenaeus bennettae[2]	.075 lb a.i./ac	8	32	29
	juvenile *Penaeus* sp	.075 lb a.i./ac	1	+++	+++

[1] Dosage expressed in lbs actual ingredient/ac from 1971 field trial: ppm from laboratory studies.
[2] Prawn data from 1970 aerial application using 1% Abate insecticide granules.

insecticides useless against mosquitoes in the Northern San Joaquin Region (Silveira, 1971). In Australia, widespread usage of insecticides for agriculture is practised but in contrast to the U.S.A., blood sucking Diptera do not warrant regular attention as a constant threat to public health because sizeable arbovirus outbreaks are so infrequent. Their pest status, however, is of some magnitude.

Granules have been applied by air previously but we know of no previous successful application at 1 lb formulation/acre. Earlier work (Standfast *et al.*, 1970) with Abate 1 percent granules at 5 lb formulation/acre indicated that a more concentrated granule would increase operating efficiency and decrease cost. For example, in 1½ to 2 hours an Agcat 300 can apply 5 percent granules at 1 lb formulation/acre to 540 acres before refueling is necessary. Insecticide cost was reduced by a half. In addition to the above advantages, at Coombabah it was found that penetration of dense mangrove canopies by the granules resulted in distribution equal to that in open country and that application could be made satisfactorily even in a 15–20 knot crosswind.

Previous work (Standfast *et al.*, 1970) established the LC_{50} of *Ae. vigilax* to Abate insecticide at 0.0006 ppm which provided some indication of the optimum treatment rate under local field conditions. During the trials being reported, larval mortality of *Ae. vigilax, C. sitiens, C. annulirostris, C. fatigans* and a few *An. annulipes* was high; those surviving treatment (apart from those hatched after flooding in the 1972 trial) were mainly pupae and late fourth instar larvae that had pupated. Abate insecticide, however, at 0.04–0.044 lb a.i./ac did not effectively control pupae although some kill was evident. The time of application, therefore is most important if maximum efficiency is to be obtained. *Aedes vigilax,* like many other *Aedes,* has eggs resistant to desiccation which are laid in salt couch and hatch when flooded by spring tides (Kerridge, 1971). Undulations within the Coombabah site caused some eggs to hatch later than others because coverage by tides was effected on different days. In 1972, the area was treated when the bulk of the *Aedes* population was at second instar. What is interpreted as increased mortality during this latter trial reflects the success of this move. However, many pupae isolated in low lying pockets within the test area still survived. Careful surveillance of breeding grounds should reveal optimum treatment days up to 10 days after a peak spring tide but one would be extremely fortunate to find a uniform population throughout. In addition, time of treatment would vary from place to place and would also be markedly influenced by recent rainfall.

Under salt-marsh conditions, Abate at the rate used has little residual effect. Used against a species exhibiting cyclic tidal emergence however, residual qualities are of little value as any insecticide left from a previous treatment would be washed away and diluted by succeeding spring tides.

Previous work has indicated relatively low hazard to non-target organisms (von Windeguth and Patterson, 1966; Mulla, 1966; Fales *et al.*, 1968; Ruber and Baskar, 1968) at low concentrations while it has been used at 0.6 ppm to control *Cyclops*, a crustacean transmitting guinea worm (Muller, 1970). At the dosages used, Abate caused no appreciable mortality of non-target organisms examined 24 hours after treatment. It was not possible in these trials to gauge the long term effects if any, of the wide scale application of this type of treatment on non-target organisms.

Granule application in mosquito control has three main advantages: (1) the most accessible life stage is being attacked (2) the potential problem of spray drift with U.L.V. techniques is nullified and (3) it can be done almost at any time of the day. Although the possibility of serious interference to natural food chains cannot at this time be dismissed, Abate 5G insecticide dispensed at 1 lb formulation/acre, should find a niche in mosquito control programmes of the future.

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SUGAR FEEDING BY FLORIDA MOSQUITOES

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Sugar is utilized by mosquitoes directly for flight energy (Hocking 1953, Nayar and Van Handel 1971) and excess amounts are converted into glycogen and fat (Van Handel 1965). Sugar is widely distributed, and mosquitoes have been recorded feeding upon flowers, extrafloral nectaries, honey-dew, and fruit juices (Smith 1904, Howard *et al.* 1912, Nielsen and Greve 1950, Hocking 1953, Haeger 1955, Nielsen and Nielsen 1958, Downes 1958, and Laarman 1968). Usually these reports are based on visual observations and only in a few cases has the presence of sugar in the mosquitoes been confirmed.

METHODS. The method used to detect sugar was based on the reaction of anthrone on fructose (Van Handel 1972). Fructose, free or as the fructose component of sucrose, is found in all plant sugars and the proportion of all fructose in a variety of Florida fruit juices and nectars ranged from 40 percent to 60 percent (Van Handel *et al.* 1972). As unfed or blood-fed mosquitoes do not contain this sugar, the presence of fructose clearly demonstrates the acquisition of a sugar meal.

Field collections were made by a suction trap (Bidlingmayer 1964), and a portable power aspirator, in a red maple (*Acer rubrum*) swamp about 10 miles west of Vero Beach. The latter method consisted of a sheet aluminum tube 13.5 inches in diameter with the distal (intake) end cut obliquely, so that the length of the tube along the long and short sides was 46 inches and 39 inches, respectively. A cloth mesh bag, the margin folded over

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