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FIELD TRIALS WITH TWO INSECT GROWTH REGULATORS AGAINST *CULEX QUINQUEFASCIATUS*

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ABSTRACT. The insect growth regulators, diflubenzuron(=Dimilin™) and Methoprene (=Altosid® 10F) applied at a target dosage of 1 ppm to larval habitats of *Culex quinquefasciatus* in a crowded section of Jakarta (trial area 1 km^2) were highly effective in preventing successful adult emergence for 2 and 5 weeks respectively after one application. Ten days after spraying with Dimilin, tarsal abnormalities were noted in emerged adults which coincided with a preponderance of emerged males.

INTRODUCTION

As part of the WHO Programme for the Evaluation and Testing of New Insecticides, the insect growth regulators Altosid® and Dimilin™ were applied to polluted breeding sites of *Culex quinquefasciatus* Say in Jakarta, Indonesia. These IGRs are also known as OMS-1697 (methoprene) and OMS-1804 (diflubenzuron), respectively. A practical evaluation of these materials under operational conditions was considered desirable because the spread of insecticide resistance to the chlorinated hydrocarbons and more recently to organophosphorus compounds in populations of this important vector are becoming serious.

DESCRIPTION OF AREA

Treatments were made in a one square kilometre area of Rawa Kerbo, a middle income crowded section in eastern Jakarta. This area is inhabited by about 20,000 people living in 4,000 houses. The main breeding sources of *Cx. quinquefasciatus* are cement and earthen drains along the roads and ground pools in back of houses. Drain width ranges from 0.3 to 0.9 metres and the water depth from 0.1 to 0.3 metres. Other common breeding sites are underground drains, while some nearby water cress fields are normally inhabited by *Cx. tritaeniorhynchus* (Giles).

The treated area was circular and a 30 ha evaluation zone, also circular, was established in the centre, and a 70 ha outer ring served as a barrier zone. Approximately 20% of the evaluation and barrier zones consisted of grassy fields and a cemetery where larviciding was not necessary. The sprayable water surface treated was approximately 2 ha, being about 2% of the total area.

Kepu, an area similar to and 4 km northwest of Rawa Kerbo, was used as an unsprayed comparison area.

TREATMENT OF AREA

The Altosid (10F formulation) and Dimilin (25% w.p.) treatments were made in September 1974 and April 1975, respectively. The target dosage applied, based on

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estimated water volume, was 1 ppm. About 2 Kg active ingredient of each material was used over the same 100 ha area, and the dosage based on the area of water surface treated was one Kg/ha, or .02 Kg/ha for the entire area.

Five spray teams (10 men) with supervisors sprayed the area with a 0.5% suspension during the morning hours, in 3 days using Hudson compression sprayers fitted with adjustable cone nozzles. The spray teams, in addition to practice spraying, made larval surveys 2 weeks earlier to familiarize themselves with breeding sites in their respective zones. A considerable amount of time was spent walking in the front and back of houses in order to locate the less obvious breeding sites in this crowded urban area.

EVALUATION METHODS

Evaluation was carried out concurrently in the sprayed and unsprayed comparison areas. Pre-treatment counts were made over a 5-day period immediately before spraying. To determine possible reductions in the natural adult population, about once a week 6 scouts sat on the verandah of houses and collected all mosquitos landing on their bared lower legs from 19.00 to 22.00 hr. Also indoor resting collections were made from 08.00 to 10.00 hr. In the laboratory, all female *Cx. quinquefasciatus* were counted, and the ovaries of unfed and freshly-fed specimens from the landing collections were removed and dried on glass slides for parous/nulliparous determination by tracheolar observation. None of the landing and resting collections was made closer than 250 m from the outer sprayed boundary.

To determine the extent of incomplete adult emergence, attempts were made weekly to collect 20 pupae from 10 fixed breeding sites. In the insectary daily observations were made of pupal mortality, incompletely emerged adults, adults that had drowned in the emergence cups and successfully emerged adults. Twenty dips were made in the same breeding sites to determine any reductions in immature density, and the average number of I-II instar larvae, III-IV instar larvae and pupae per dip were recorded.

To determine whether mortality occurred at a post-treatment moult, on 2 occasions several hundred 2nd instar larval specimens were collected from the Dimilin treated drains and observed in the laboratory for 1 wk.

Limited observations also were made on the life span of emerged adults in the Dimilin trial area. Prior to spraying and also 10 and 20 days after spraying, special collections were made to obtain pupae from the field. Both sexes were allowed to emerge in the same cages (ca. 25 cm^3) which were kept draped with wet towels, and the adults were provided with cotton wool soaked in 5% sugar solution. Each day, any dead mosquitoes were removed and counted. The mean minimum and maximum temperatures in the insectary were 27.1°C and 30.6°C, respectively. The mean relative humidity was 77%.

RESULTS

The female landing population after Altosid treatment was reduced by 85% from the 2nd until the 5th week after spraying (Table 1). This figure is based on an average pre-control density of 11.5 versus 1.8 for the above interval after spraying. The period of control with Dimilin was shorter, the average pre-control density of 3.1 being reduced by 54% up to 10 days after spraying. In both treatments the parous rate nearly doubled, indicating that emergence within the treated area had been temporarily suppressed. A higher density and lower (but stable) parous rate occurred in the unsprayed area during both spray operations.

With Altosid, the indoor resting reduction was very similar to that of the landing rate reduction. For 1 wk after treatment the resting density in houses was essentially unchanged, being 24 females per man hour. Then, from wk 2 to wk 6 it averaged 67% below the pre-treatment mean. While even after 6 months the rest-

Table 1. Density and proportion parous of female *Cx. quinquefasciatus* adults after IGR treatment. In parentheses are numbers dissected. Six scouts collected mosquitoes attracted to their lower legs from 19.00 to 22.00 hours.

Day after treatment with one ppm	Altosid trial				Dimilin trial			
	Sprayed		Unsprayed		Sprayed		Unsprayed	
	No. per man hr	Prop. parous	No. per man hr	Prop. parous	No. per man hr	Prop. parous	No. per man hr	Prop. parous
—	13.5	.36 (53)	—	—	2.3	.18 (22)	15.2	.34 (99)
—	9.6	.32 (77)	—	—	3.8	.51 (57)	11.2	.34 (90)
2	20.2	.29 (95)	—	—	—	—	—	—
3	—	—	—	—	1.8	.58 (31)	—	—
6	—	—	—	—	1.4	.33 (18)	—	—
7	14.1	.51 (83)	—	—	—	—	12.0	.29 (99)
8	—	—	—	—	1.3	.67 (21)	—	—
10	—	—	—	—	1.2	.57 (16)	—	—
13	—	—	—	—	1.9	.52 (31)	—	—
14	—	—	—	—	—	—	8.2	.24 (96)
15	3.7	.52 (48)	—	—	3.1	.44 (41)	—	—
16	—	—	25.1	.26 (98)	—	—	—	—
17	—	—	—	—	2.2	.27 (30)	—	—
21	1.1	(0)	—	—	2.2	.50 (36)	—	—
28	0.6	(0)	—	—	3.1	.33 (46)	—	—
31	—	—	30.4	.34 (92)	—	—	—	—
35	1.9	.62 (29)	—	—	—	—	—	—
36	—	—	19.6	.25 (20)	—	—	—	—
42	6.1	.48 (77)	—	—	—	—	—	—
49	6.6	.49 (76)	—	—	—	—	—	—
52	—	—	13.2	.28 (80)	—	—	—	—

ing density was at least 35% below the pretreatment level, this was probably due to seasonal change. With Dimilin, no appreciable reduction in resting density occurred, which was 12 females per man hour before spraying.

Observations on the extent of incomplete adult emergence indicated that few pupae were killed in the Dimilin trial and that mortality was more apparent in emerging adults (Table 2). However, relatively few pupae were found during the first 2 wk after spraying, indicating considerable larval mortality. From 17 to 27 days after spraying, pupal numbers increased, and 36% of them produced adults which were unable to successfully emerge. With Altosid, during the first 3 wk after spraying nearly all mortality occurred during the pupal stage with very few adults emerging (Table 3). Thereafter, even though pupal mortality gradually decreased, significant mortality occurred among adults, both incompletely and completely emerged, until finally there was more adult than pupal mortality.

With Dimilin, for all immature stages, a 78% reduction in density occurred for up to 15 days after spraying (Table 4). But pupal counts continued to be reduced by 60% for an additional 2 wk. Eight days after spraying a 96% reduction in pupae occurred. With Altosid, attempts to analyze data for larvae and pupae suffered from the usual problem of high variance among dips and among days. There was no significant reduction in immature density when compared to the unsprayed comparison area. During the first 35 days after spraying, the mean numbers per dip of I-II instar larvae, III-IV instar

Table 2. Inhibition of adult emergence of *Cx. quinquefasciatus* after Dimilin treatment. Pupae obtained by dipping in 10 fixed breeding sites.

Day after treatment with one ppm	Number of pupae observed	Percentage mortality: Pupae	Incompletely emerged adults	Completely emerged adults	Total
		Sprayed			
—	215	3	0	1	4
—	210	6	0	1	7
—	167	6	0	2	8
2	31	20	0	35	55
8	2	100	0	100	100
10	41	0	0	46	46
15	58	12	0	28	40
17	155	11	0	39	50
20	83	6	0	49	55
23	120	8	0	31	39
27	114	11	0	25	36
		Unsprayed			
—	80	5	0	3	8
—	155	5	1	1	7
—	110	5	0	0	5
1	146	3	0	1	4
9	95	3	1	1	5
14	130	3	0	3	6
22	168	2	0	0	2

larvae and pupae in the Altosid area were 14.3, 13.0 and 7.9, respectively.

The second instar larvae collected from Dimilin treated sites showed high mortality either before or after they moulted to third instars. Total mortality 6 days after spraying was 99%, but it was only 23% after 10 days.

Observations on the life span of successfully emerged adults from the Dimilin trial area showed little difference in the longevity of specimens obtained before and 20 days after spraying. About 50% of both sexes had died by day 20, although a few specimens were still alive by day 40. However, the small number of adults (19 ♀ 53♂) which had emerged from pupae collected 10 days after spraying exhibited high mortality on the third and fourth day after emergence and no specimen lived longer than 18 days. Many rested on the cage floor, after emergence from the pupal skin and flight from the water surface. The front legs of these mosquitoes were bent upwards at the third tarsal segment and, although the wings appeared normal and active, they could not maintain proper balance to lift off and fly despite escaping from the small water containers used for holding pupae.

Further observations on the above experiment with Dimilin showed that only 24% of the emerged adults (228 specimens) derived from pupae 10 and 20 days after spraying were females, as opposed to 42% (173) before spraying and 56% (235) in the unsprayed area.

DISCUSSION

One application of Altosid and Dimilin at a high dosage rate was highly effective in preventing successful adult emergence of *Cx. quinquefasciatus* in Jakarta. Altosid

Table 3. Inhibition of adult emergence of *Cx. quinquefasciatus* after Altosid treatment. Pupae obtained by dipping in 10 fixed breeding sites.

Day after treatment with one ppm	Number of pupae observed	Percentage mortality: Pupae	Incompletely emerged adults	Completely emerged adults	Total
			Sprayed		
—	157	6	1	0	7
—	166	2	1	0	2
—	159	2	0	0	2
2	199	79	3	0	81
8	164	99	0	0	99
10	162	100	0	0	100
17	149	96	1	1	99
21	122	89	3	6	97
28	140	75	7	4	86
35	117	39	11	10	61
42	100	33	9	9	51
53	147	17	6	25	48
60	77	7	0	9	16
			Unsprayed		
—	153	3	1	0	5
—	160	2	4	0	7
4	152	3	0	0	3
9	159	3	1	0	4
16	158	3	3	0	5
23	142	2	1	0	3
30	148	1	1	0	2
36	142	4	1	0	4
44	165	5	0	0	5
65	159	2	1	0	3

appeared to be more persistent and also caused a higher reduction in the natural adult population. The shorter persistence of Dimilin probably permitted appreciable adult emergence before some adults already present before spraying had died. Both materials probably would have caused greater adult reductions if the area under control had been larger thus reducing infiltration from unsprayed areas.

Low dosages (.028 Kg/ha) of Altosid and Dimilin have been effective in aircraft applications against *Aedes nigromaculis* and other species in pastures in California (Schaefer et al. 1974, 1975). A second application is normally required about 10 days later if the next brood is to be prevented. Target field dosages lower than one ppm should be considered in any future trial in Indonesia. With a view to reducing costs, operational and economical factors to consider are increased labor costs necessary to carry out repeated applications of a less persistent dosage as compared to the fewer applications and lower labor costs associated with a dosage providing more persistent action.

Mulla and Darwazeh (1976) recently reported that Dimlin had great potential for rendering long-lasting control of asynchronous populations of immature mosquitoes at very low practical dosages.

Table 4. Density of *Cx. quinquefasciatus* larvae and pupae after Dimilin treatment. Twenty dips were made in 10 fixed breeding sites. No reduction in immature density occurred in the Altosid trial.

Day after treatment with one ppm	Average number per dip		
	I–II instars	III–IV instars	Pupae
		Sprayed	
—	8.6	4.2	1.8
—	11.5	3.8	2.2
2	0.5	1.9	0.1
8	2.8	1.5	0.08
15	1.3	1.1	0.7
20	10.5	4.4	0.7
27	10.1	5.1	0.9
		Unsprayed	
—	20.8	10.0	2.5
—	30.5	35.4	15.7
2	3.9	5.5	4.1
8	9.7	6.5	2.6
15	18.2	15.3	8.0
29	15.1	21.0	12.8

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