A field evaluation study on the effects of residual spray of Bifenthrin and Deltamethrin on *Anopheles minimus* population in Mae Hong Son Province, northern Thailand.

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Abstract: A field study was conducted to evaluate the effects of indoor residual house spraying of Bifenthrin and Deltamethrin on malaria vector population of *Anopheles minimus* s.l., from April 1999 to April 2001 at rural villages in Mae Hong Son province, northern Thailand. Nine villages in Mae Hong Son province were selected for the present study (three villages for control and three villages each for insecticide spray). The residual spray of Bifenthrin (25 mg/m^2) showed greater adulticiding effects on *An. minimus* s.l. population than Deltamethrin (20 mg/m^2). In Bifenthrin treated villages, a clear decrease in biting density of *An. minimus* s. l. was found in human bait collection as well as animal bait collection after the insecticide spray. In all of the three villages, the average density after insecticide spray was significantly lower than that observed before the spray. The effects of Deltamethrin on *An. minimus* s. l. density was found only in one village out of the three treated villages. A significant decrease in parous rate after insecticide spray was found in all the villages sprayed with Bifenthrin, whereas no significant changes were observed in control villages. The average parous rate in the villages treated with Deltamethrin became significantly higher after the insecticide spray. These results clearly suggested that the residual spray of Bifenthrin (25 mg/m^2) was more effective than Deltamethrin (20 mg/m^2).

Key words: residual spray, Bifenthrin, Deltamethrin, *Anopheles minimus*, Thailand

INTRODUCTION

The malaria control program in Thailand was started in 1950's and has resulted in an impressive reduction in malaria morbidity and mortality. For more than 40 years, DDT has been used in the malaria control program for indoor residual spraying throughout Thailand. However, the use of DDT for malaria control in Thailand has been decreasing from 1980's and nearly stopped recently because of the development of insecticide resistance in vector populations against DDT and the side effects of DDT spraying on the environment through the biological concentration (Malikul, 1988; Curtis, 1994; Chareonviriyahpop *et al.,* 1999).

The screening of insecticide alternative to DDT has become an important subject for malaria control in Thailand, since insecticide spraying is still the most effective control measure to stop the malaria transmission, especially during its epidemic. Several field studies have been carried out recently in Thailand on the effectiveness of insecticide in malaria vector control (Photijitthi *et. al.,* 1999; Prajakwong *et. al.,* 1997ab; Somboon *et. al.,* 1995; Vongprayoon *et. al.,* 1999, Suwonkerd *et. al,* 1997). Bifenthrin is a newly synthesized pyrethroid and the effectiveness has been studied preliminary in the Office of Vector Borne Diseases Control 2 (VBDO2), Chiangmai, Thailand in 1998. Based on the results of the preliminary study, the present study was conducted to compare the effects of indoor residual house spraying of Bifenthrin and Deltamethrin on malaria vector population of *Anopheles minimus* s.l., from April 1999 to April 2001 at rural villages in Mae Hong Son province, northern Thailand.

MATERIALS AND METHODS

Study area: Mae Hong Son province is located at the Thai-Myanmar border in northwestern Thailand, and the incidence of malaria in the province is the highest in the upper northern part of Thailand (VBDO2, 1999). Nine villages in Mae Hong Son province were selected for the present study. The total number of houses, population, percent-

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age of houses having a bed net and average surface area to be sprayed in each village were shown in Table 1. Three villages each were sprayed with insecticide (Bifenthrin or Deltamethrin) and remaining 3 villages were left without insecticide spraying (control villages).

Insecticide application: Insecticide was applied two times in April and August 2000. Bifenthrin 25 mg/m^2 or Deltamethrin 20 mg/m^2 was applied to the inside wall of household by using Hudson X-pert hand compression pumps with flat fan 8002 nozzles. The actual dosage of insecticides applied on the wall was checked by WHO (Head Quarter in Geneva, Switzerland) by processing the filter papers (Whatman No.1) placed on a wall before the insecticide spraying.

Evaluation of the effect of insecticide spray on vector mosquitoes: The following two different methods were used for evaluating the effects of insecticide spray on vector mosquitoes; (1) survey of wild vector populations and (2) surface contact bioassay.

The wild vector population was surveyed monthly in each village by using an indoor and outdoor human bait collection and animal bait collection. Four houses in each village were selected and a pair of collector sat inside and outside of the houses. Landing mosquitoes were collected for 50 min with a 10 min of break interval from 18:00 to 24:00. For the animal bait collection a cow was tethered inside a gauze net (4 by 4 by 2 m), which was similar to the one described by Service (1993). One collector aspirated mosquitoes landing in and out of the net at every 15 min from 18:00 to 24:00. The collected mosquitoes were kept in a plastic cup hourly for the later identification. During the mosquito collection relative humidity and temperature were recorded. Among female *An. minimus* s. l. collected from human bait collection, at least 50 unfed females were dissected for the parity check (Detinova, 1962) and were checked for malaria sporozoite in salivary glands.

The surface contact bioassay test (WHO, 1975) was carried out monthly after the1st insecticide application. One house in the sprayed villages was selected for the test. Five different places on the inside wall was marked and used for the insecticide exposure test. Twenty females of 3-5 day-old *An. minimus* s. l. laboratory strain (Mae Hong Son strain) were aspirated into each cone, and totally 100 females were used for each test. The females were exposed to sprayed surface for 30 min, and the number of dead and knocked down mosquitoes were counted at the end of exposure. The number was counted again after 24 hr. Abbot's correction was applied to calculate mortality rate based on the mortality rate of control group.

Data analysis: The average density of *An. minimus* s. l. was calculated for two periods, before and after the insecticide spraying, and the significance of the difference was tested by the t-test. The difference in parous rate was analyzed by ANOVA or the t-test after made the arcsine transformation. For the statistical analysis of the surface contact bioassay test, the study period was divided into 3 periods of 4 month, and the average mortality was calculated and compared after the arcsine transformation. Variation in temperature and humidity among village was analyzed by ANOVA, and Tukey's HSD test was used for a pair wise comparison of means. All statistical analysis was performed by using Systat statistical software (Wilkinson, 1996).

RESULTS AND DISCUSSION

Coverage of indoor insecticide spraying and climate conditions during the study: The percentage of houses sprayed with insecticide in this study was shown in Table 1. The coverage of insecticide spray reached more than 89% of the total houses, except for one village, Pong Kan Nai where the coverage was 79.1 and 80.6% at1st and2nd insecticide spray, respectively. Houses without resident or refused by house owner were not treated with insecticide.

Significant differences in humidity were observed among the study villages ($F=5.401$, $p<0.001$, Table 1). The average humidity recorded in the villages selected for Bifenthrin spray were significantly lower than other villages, while no significant differences were found for temperature among villages ($F=1.116$, $p=0.353$).

Effects of insecticide application on wild population of* An. minimus *s. l.: A total of 80,662 anopheline of 22 species were collected in the present study. Among them 30,340 and 98 were morphologically identified as A*n. minimus* s.l. and *An. dirus* s.l. respectively, clearly showing that *An. minimus* s.l. was the most important malaria vector in the study villages. A total of 12,600 *An. minimus* s.l. were dissected for the detection of malaria sporozoite in the salivary gland and all of them were negative.

Table 2 compares the average density of *An. minimus* s. l. before and after the insecticide spray. The residual spray of Bifenthrin showed greater effects on *An. minimus* s.l. population than Deltamethrin. The seasonal changes in *An. minimus* s.l density observed before the insecticide spray was similar to that reported in previous papers (Ismail *et al.,* 1974; Ismail *et al.,* 1975; Suwonkerd *et al.,* 1995; Takagi *et al.,* 1995; Suwonkerd *et al.,* 1997). However, in Bifenthrin treated villages a clear decrease in biting density of *An. minimus* s. l. was found in human bait collection as well as animal bait collection after the insecticide spray. In all of the three villages, the average density after the insecticide spray was significantly lower than that before the spray (t-test, $p<0.05$).

Table 1. Demographic parameters, climate conditions and results of insecticide application at each study village in Mae Hong Son, northern Thailand.

	Village								
	Tong Muang	Pakolo	Huey Pong On	Pong Kan Nai	Sao Tao	Chang Chum	Mai Sape	Huey San Nok	Tobsok
Number of houses	49	52	82	61	136	101	92	36	74
Population	141	181	304	326	556	410	426	119	276
Average (±sd) temperature during the study*	25.0^{a}±4.0	24.3^{a}±3.4	23.6^{a}±4.8	23.9^{a}±4.1	25.3^{a}±3.1	23.4^{a}±3.4	23.1^{a}±3.9	24.7^{a}±3.2	25.2^{a}±2.7
Average (±sd) humidity during the study*	68.8^{bc}±8.8	68.8^{c}±8.2	67.6^{c}±7.8	75.8^{ab}±13.3	77.9^{ab}±9.6	79.1^{ab}±9.1	76.2^{ab}±7.3	77.5^{ab}±6.6	80.2^{a}±17.3
% houses having a bed net	93.8	98.0	85.2	100	87.5	93	93.5	86.1	87.8
Average surface to be sprayed (m^2)/house	163.8	206.2	175.6	198.9	280.9	266.9	184.8	247.8	223.8
Insecticide sprayed	Bifenthrin			Deltamethrin			No spray		
% houses sprayed**									
1st application	92.6	96.4	90.2	79.1	91.6	94.9	—	—	—
2nd application	92.3	93.3	89.4	80.6	92.8	92.8	—	—	—

*Averages in the same row followed by the same letter were not significantly different (Tukey's HSD test, p>0.05).
**1st application = 24-27 April 2000; 2nd application = 23-25 August 2000.

Table 2. Comparisons of average density (±sd) of *An. minimus* s. l. observed before and after the insecticide spray in Mae Hong Son, northern Thailand from April 1999 to April 2001.

Village	Indoor Collection			Outdoor Collection			Animal bait Collection		
	Before spray	After spray	p	Before spray	After spray	p	Before spray	After spray	p
Bifenthrin sprayed village									
Tong Muang	43.1 ±34.7	3.5 ±3.5	<0.001	73.7 ±40.4	18.9 ±11.0	<0.001	10.1 ±13.0	0.3 ±0.7	0.004
Pakolo	84.8 ±74.1	11.3 ±9.6	<0.001	183.3 ±150.6	82.4 ±88.6	0.039	37.2 ±54.1	2.9 ±8.1	0.012
Hueypong On	18.5 ±18.6	0.8 ±1.4	<0.001	43.0 ±43.0	11.1 ±11.9	0.006	15.4 ±26.7	0	—
Deltamethrin sprayed village									
Pong Kan Nai	68.7 ±87.9	91.4 ±162.6	0.719	176.6 ±211.7	193.8 ±254.7	0.870	33.3 ±45.4	39.5 ±36.4	0.711
Sao Tao	32.1 ±24.9	5.0 ±3.5	<0.001	67.6 ±44.8	22.6 ±12.4	<0.001	0.3 ±0.9	7.8 ±8.0	0.034
Chang Chum	68.6 ±56.8	47.8 ±67.8	0.458	168.1 ±133.7	113.5 ±114.6	0.296	3.2 ±3.9	29.5 ±35.8	0.076
Control village									
Tob Sok	8.0 ±8.8	14.3 ±6.7	0.069	38.7 ±39.4	33.9 ±18.7	0.689	No data	0	—
Mai Sa Pe	2.0 ±3.7	8.62 ±11.0	0.141	8.8 ±13.6	22.8 ±40.2	0.377	1.2 ±1.6	1.0 ±2.4	0.876
Huey San Nok	10.8 ±16.5	62.4 ±53.8	<0.001	47.7 ±53.3	178.6 ±130.5	0.025	1.7 ±4.6	0	—

*The insecticide was sprayed twice; 1st spray= April 2000, 2nd spray= August 2000.

The effects of Deltamethrin on *An. minimus* s. l. density was found only in one village (Sao Tao) out of three villages. The average biting density on human bait was significantly reduced after the insecticide spray, whereas in animal bait collection it showed significant increase after the insecticide spray (t-test, p<0.05).

No significant increase or decrease was found in biting density of *An. minimus* s. l. in control villages.

The average parous rate of mosquitoes was calculated for each village and compared before and after the insecticide spray in Table 3. A significant decrease in parous rate after insecticide spray was found in all the villages sprayed with Bifenthrin, whereas no significant changes were observed in the control villages. The average parous rate in the

Table 3. Comparisons of average (±sd) parous rate before and after the insecticide spray in Mae Hong Son, northern Thailand from April 1999 to April 2001.

	Before spray	After spray	p
Bifenthrin sprayed village			
Tong Muang	70.8 ±13.5	32.3 ±11.2	<0.001
Pakolo	69.3 ±16.7	32.7 ±16.0	<0.001
Hueypong On	72.6 ±12.7	31.7 ±13.5	<0.001
Deltamethrin sprayed village			
Pong Kan Nai	47.3 ±17.5	78.5 ±13.2	0.005
Sao Tao	50.0 ±18.4	84.0 ±6.3	<0.001
Chang Chum	43.3 ±17.6	73.1 ±19.1	0.020
Control village			
Tob Sok	38.9 ±16.3	46.9 ±18.4	0.383
Mai Sa Pe	37.2 ±22.1	44.9 ±18.5	0.624
Huey San Nok	41.5 ±16.3	49.1 ±19.3	0.392

Table 4. Average mortality (±sd) of adult *An. minimus* s. l. observed by the bioassay test in 3 different month periods after the insecticide spray*.

	Period after 1st spray (month)	Bifenthrin	Deltamethrin	p
1st spray				
	1-4	98.50 ±2.00	77.50 ±15.92	<0.001
2nd spray				
	5-8	97.75 ±2.91	79.75 ±8.94	<0.001
	9-12	76.75 ±12.24	72.75 ±32.01	0.674

*The insecticide was sprayed twice; 1st spray= April 2000, 2nd spray= August 2000.

villages treated with Deltamethrin became significantly higher after the insecticide spray. No significant decrease was observed in parous rate as well as the biting density of *An. minimus* s.l. in a village treated with 25 mg/m^2 of Deltamethrin in a previous study (Prajakwong *et al.,* 1997a).

Results of surface contact bioassay test: The mortality of adults exposed to the Bifenthrin-treated wall was always higher than that found on Deltamethrin-treated wall (Table 4). The difference in mortality between Bifenthrin and Deltamethrin was significant during the 1-4 month and 5-8 month, but not during the 9-12 month (Tukey's HSD test). The average mortality of adults exposed to a Bifenthrin-treated wall during the 1-4 month and the 5-8 month was 98.5 and 97.75, respectively, and the difference was not significant (t-test, p>0.05). However, the mortality decreased significantly to 76.75 % in the 9-12 month. On the Deltamethrin-treated wall the adult mortality was less than 80% and not significantly different between the 3 study periods (F=0.019, p=0.981), and within the first 4 months after spray, the mortality of exposed adults decreased rapidly from 92.0 to 67 %. With a higher dosage of Deltamethrin (25 mg/m^2) than the present study (20 mg/m^2), Prajakwong *et al* . (1997a) observed the mortality rate of exposed adult *An. minimus* s. l. from 100-92 % during the first 3 months. The lower residual effects of Deltamethrin, because of the low dosage in this study, might be one of the reasons for the less effectiveness of Deltamethrin to *An. minimus* s.l. population.

These results clearly suggested that the residual spray of Bifenthrin (25 mg/m^2) was more effective than Deltamethrin (20 mg/m^2). However, the average density of *An. minimus* s.l. achieved in Bifenthrin-treated villages was nearly the same level as that observed in the control villages (Table 2). Since indigenous malaria cases, in which the infection was acquired inside the village of residence, were reported every year from the control villages, a higher dosage or more frequent spray of the insecticide is required to control malaria from in these villages.

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