Movement of individually marked *Aedes albopictus* females in Nagasaki, Japan.

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Abstract: Sequential movement of *Aedes albopictus* was observed by the release of individually marked females in Nagasaki, Japan. Three cohorts of laboratory females were individually marked and released for 3 successive days from a point in an isolated, small, grassy and scrub area in May, 1990. Human bait collection was made at 10 fixed collection points for 15 days, and collected females were checked for the mark. The marked females were released again from the point of recapture. Unmarked females collected were individually marked and released from the point where they were collected. From the repeated recapture data, sequential movements of females among 10 collection points were recorded and analyzed. Some points characterized by accumulation and/or frequent exchange of females were distinguished through the analysis. The spatial variation in the number of biting females among 10 collection points was partly explained by repeated movements among collection points.

Key words: individual marking, movement, *Aedes albopictus*, Nagasaki

INTRODUCTION

Behavioral studies of biting mosquitoes are required to estimate the degree of vector-human contact, which is one of the important parameters relevant to vectorial capacity of mosquitoes. Individual marking is one of the possible methods to study the behavior of biting mosquitoes analytically.

Although many mark-release-recapture studies have been made (Service, 1993), individual marking of mosquitoes has been used only in a few studies on ecology of *Aedes aegypti* (Sheppard *et al.*, 1969; Trpis and Hausermann, 1986). Sheppard *et al.* (1969) estimated population parameters such as survival rate, population density, daily recruitment, and flight distance, in relation to dengue haemorrhagic fever in Bangkok. Dispersal of *Ae. aegypti* was studied by Trpis and Hausermann (1986) in Kenya using individual marking method. Besides the estimation of population parameters, they showed movement of mosquitoes among human houses through the analysis of sequential movements of marked mosquitoes. The

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usefulness of individual marking in behavioral study of biting mosquitoes has been shown in these studies.

Using *Ae. albopictus,* several mark-release-recapture studies have been made in Japan (Mori, 1979; Mori and Wada, 1977; Takagi *et al.*, 1995a, b) and USA (Bonnet and Worcester, 1946; Niebylski and Craig, 1994). Not only the flight distance but also behavioral aspects of released mosquitoes were examined in these studies, however no individual marking studies were made to analyze details of movement within a study area (Hawley, 1988).

The sequential movements of biting mosquitoes within a small area were observed by individual marking of *Ae. albopictus* in the present study. Through analysis of the sequential movements, spatial variation in the number of mosquitoes among collection points was discussed.

MATERIALS AND METHODS

The study was made in an isolated, small, grassy and scrub area (0.8ha) known as Gubirogaoka on the campus of Nagasaki University School of Medicine. Three types of vegetation were noted in the area (Fig. 1). TG area had a dense vegetation of trees, shrubs, and herbaceous plants. Dominant tree species included *Cinnamomum camphora, Ilex integra,* and *I. crenata*. SG area was covered with shrubs and herbaceous plants. G area had a direct sun shine and was covered with herbaceous plants. *Aedes albopictus* was the dominant mosquito species in the study area.

Unfed females of laboratory strain of 4−5 days after emergence were marked individually and released for 3 successive days (22−24 May, 1990). Three cohorts of 296, 161, and 249 females were released from a single point (No. 22) on the 1st, 2nd, and 3rd day of

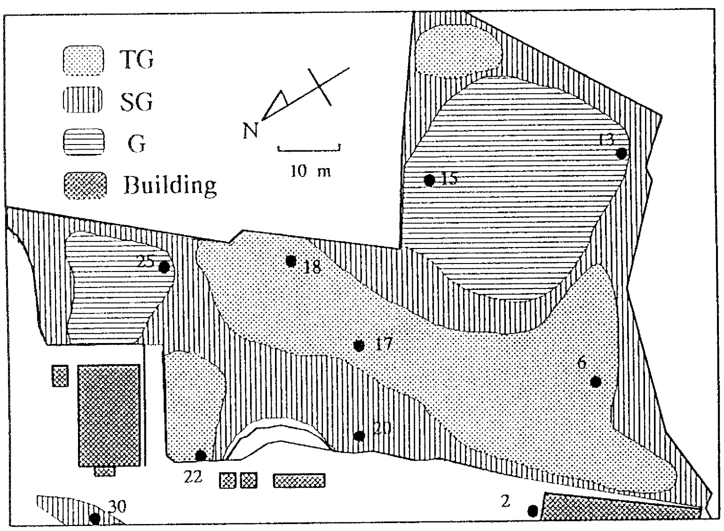

Fig. 1 A map of the study area showing vegetation and 10 collection points of *Ae. albopictus* females.

the experiment, respectively. A total of 150 unfed females from the field population was also marked and released from the point where they were collected during the experiment. Human bait collections were made at 10 fixed collection points from 23 May to 6 June, 1990. Because of heavy rain the adult collection was not made on 1 and 4 June. One person carrying 500g of dry ice collected females landing on his exposed legs by glass vials (2cm in diameter and 5cm high) for 7 min. All the collected females were checked for the mark in the laboratory. Marked females were returned within 2hrs to the point where they were collected, and unmarked ones were returned after individual marking. The marking of unmarked females from the field population was continued throughout the study period.

To make individual mark, mosquitoes were anesthetized by carbon dioxide gas and placed on a nylon mesh attached to the end of a plastic pipe (7cm in diameter) which was connected to an vacuum cleaner. When marked, they were sucked gently by the cleaner and their wings were fixed on the nylon mesh. A felt tip pen was used to mark 6 different positions of the wings (Fig. 2). By placing single color dots of 3 different colors in a binary notation, as many mosquitoes as $4^6-1=4095$ could be differently marked.

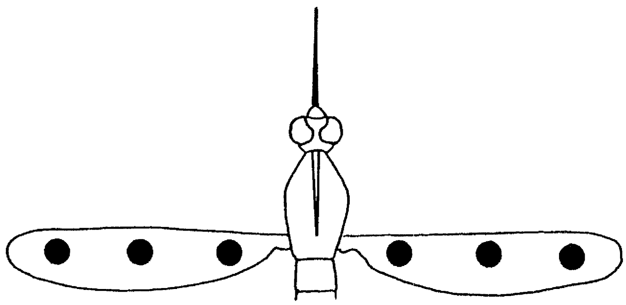

Fig. 2 Diagram showing the 6 positions on the wings of mosquito used for marking.

Result and Discussion

Among 856 released females, a total of 116 (13.0%) females was collected at least once and 21 (2.5%) of them were collected twice (Table 1).

The daily changes in the number of recaptured mosquitoes of 3 laboratory cohorts are shown in Table 2. A few females of cohorts 1 and 2 were recaptured on the 1st and 2nd days postrelease. On 26 and 27 May, the numbers of recaptured females became small in all the cohorts. The small numbers of recaptured females during 31 May to 4 June were ascribed to rain. The effects of weather were not negligible in the present study, therefore, it was difficult to apply logregression analysis to estimate the daily loss rate of released females under the assumption of constant mortality and emigration rates during the experiment.

The interval between the 1st and the 2nd recapture of marked females is shown in Table 3 for 20 females from laboratory cohorts. The average days of the 1st recapture after

Table 1. Numbers of released females and females recaptured at least once and twice.

	Total released	The 1st recapture	The 2nd recapture
Cohort 1	296	55	11
Cohort 2	161	17	3
Cohort 3	249	20	6
Field population	150	24	1
Total	856	116	21

Table 2. Daily change in the number of recaptured mosquitoes of 3 laboratory cohorts.

	May								June						
	23	24	25	26	27	28	29	30	31	1	2	3	4	5	6	Total
Cohort 1	1	4	14	2	1	12	5	6	2	—	1	3	—	0	4	55
Cohort 2		1	2	0	0	6	3	3	1	—	0	0	—	1	0	17
Cohort 3			7	0	1	3	2	5	1	—	1	0	—	0	0	20
Total	1	5	23	2	2	21	10	14	4	—	2	3	—	1	4	92

Table 3. Interval between the 1st and the 2nd recapture of marked females in relation to days of the 1st recapture.

Days of the 1st recapture	Days to the 2nd recapture							
	1	2	3	4	5	6	7	8
1			1	1				1
2	1				1			
3		1		1				
4		1	2					
5		2						
6	1	1			1			
7	2							
8	1							
9								
10								
11								
12	2							

the release and the average interval between the 1st and the 2nd recapture were calculated as 4.95±3.20 and 2.60±1.85, respectively. Females recaptured at 6−12 days post-release came to bite again on the next day. Mori and Wada (1977) released marked *Ae. albopictus* and recaptured parous females at 4−10 days post-release. Takagi *at al.* (1995a) released fed *Ae. albopictus* and observed significant increased of egg numbers in ovitraps at 4−7 days after blood feeding. Taking account of the lower average temperature of 20.7℃ in the present study, it is likely that females recaptured at 6−12 days post-release had finished the 1st or 2nd gonotrophic cycle and were actively seeking for blood meal.

The sequential movements of 20 and 24 recaptured females from the laboratory cohorts and the field population, respectively, were analyzed. As to the laboratory cohorts, the sequential movements of females which were recaptured twice were used in the analysis. The movements of released mosquitoes among collection points were summarized in Table 4 through the same analysis as Trpis and Hausermann (1986). The column of Table 4 shows the next recapture point of females which had been recaptured at the same point in the previous collection. For example, 23 females had been collected and released from No. 22 in the previous collection, and 9 of them were collected again at No. 22. Among the other females, 8, 2, 2, 1, and 1 diverged from No. 22 to No. 18, 25, 17, 30, and 6, respectively. The total number of diverging females was counted in Table 5 as the frequency of divergence (*D*) from each point.

The row of Table 4 shows the previous recapture point of females which were collected at the same points in the 2nd recapture. Among 14 females collected at No. 22 in the 2nd recapture, 4 and 1 converged from No. 30 and 17, respectively. The total number of

Table 4. Movements of mosquitoes among 10 collection points.

Next recap. point	Previous recapture point										Total
	22	18	25	13	15	20	17	30	6	2	
22	9						1	4			14
18	8	3			1						12
25	2		3					1			6
13											0
15				1							1
20			1				1				2
17	2	4				1					7
30	1					1					2
6	1										1
2											0
Total	23	7	4	1	1	2	2	5	0	0	45

converging females was counted in Table 5 as the frequency of convergence (*C*) to each point. The females collected at the point of previous release were either staying at or going out and returning to the previous point, and thus, distinguished from diverging or converging females, and the total number was shown in Table 5 as the frequency of "stay or return".

The females of "stay or return" were observed only in 3 collection points, No. 22, 18, and 25. The higher frequency of "stay or return" may suggest the higher probability of staying and returning, and leading to accumulation of females around the point. The balance between convergence and divergence ($C-D$) was calculated and shown in Table 5. We can expect decrease and increase of density at each collection point from the negative and positive sign of the balance, respectively. The total number of movement ($C+D$) shows the frequency of movement around the collection point. In collection points No. 22 and 18, movements were observed more frequently than other collection points. The positive sign of the balance and 3 females of "stay or return" both in No. 18 and 25 suggest that females were accumulated around these collection points. The negative balance and the highest frequency of "stay or return" were observed in No. 22. In Table 4 "both-way" movements were observed between No. 22−17, No. 22−30, and No. 17−20. As shown in Fig. 1, the distances among these 4 points were short and 3 or the points located at the edge of vegetation, and this situation may allow frequent movement of biting females among these points. Takagi *et al.* (1995b) released marked females of *Ae. albopictus* from No. 22 and showed that 63.7% of recaptured females were collected in No. 17, 18, and 25 around the release point. Their analysis suggested that the small distance among these collection points and environmental conditions such as the vegetation structure are the main reason of a large number of recaptures in these points.

The densities of unmarked females (UMK in Table 5) at No. 22, 18, and 25 were high as expected from the present analysis of movement, but it was difficult to explain the density of biting females at other points. For example, the sign of the balance in No. 17 was positive

Table 5. Summary of movements among 10 collection points.

	Collection point									
	22	18	25	30	20	17	15	13	6	2
Convergence (C)	5	9	3	2	2	7	1	0	1	0
Divergence (D)	14	4	1	5	2	2	1	1	0	0
Stay or return	9	3	3	0	0	0	0	0	0	0
C+D	19	13	4	7	4	9	2	1	1	0
C−D	−9	5	2	−3	0	5	0	−1	1	0
UMK*	49	39	29	21	19	12	19	23	10	7

*Total number of unmarked females

and a high density was expected, but the observed number of biting females was small. The density of biting mosquitoes is determined by not only movement (emigration and immigration) but also mortality and recruitment of adults. Therefore, the movement of biting females shown in the present analysis partly explains the spatial variation in the number of unmarked females. Other factors relating to mortality and recruitment of females are also important to understand the spatial variation in the number of biting females.

The present study shows that individual marking is a useful method to analyze movements of biting females among multiple collection points. But the handling procedures of individual marking are complicate and may have some harmful effects on behavior and survival of marked mosquitoes. The small number of recaptured females just after the release in the present study may be partly because of the effects of our handling procedures. To establish more convenient and harmless procedures, some experiments will be needed in the future study.

REFERENCES

1) Bonnet, D.D. & Worcester, D.J. (1946): The dispersal of *Aedes albopictus* in the territory of Hawaii. Am. J. Trop. Med., 26: 465–476.
2) Hawley, W.A. (1988) The biology of *Aedes albopictus*. J. Am. Mosq. Control Assoc. (Suppl.) 1: 1–40.
3) Mori, A. (1979): Effects of larval density and nutrition on some attributes of immature and adult *Aedes albopictus*. Trop. Med., 21: 85–103.
4) Mori, A. & Wada, Y. (1977): The gonotrophic cycle of *Aedes albopictus* in the field. Trop. Med., 19: 141–146.
5) Niebylski, M.L. & Craig, Jr, G.B. (1994): Dispersal and survival of *Aedes albopictus* at a scrap tire yard in Missouri. J. Am. Mosq. Control Assoc., 10: 339–343.
6) Service, W.W. (1993): Mosquito Ecology, Field Sampling Methods, 2nd ed., 988pp. Elsevier Applied Science, London.
7) Sheppard, P.M., Macdonald, W.W., Tonn, R.J. & Grab, B. (1969): The dynamics of an adult population of *Aedes aegypti* in relation to dengue haemorrhagic fever in Bangkok. J. Anim. Ecol., 38: 661–702.
8) Takagi, M., Tsuda, Y. & Wada, Y. (1995a): Movement and oviposition of released *Aedes albopictus* (Diptera: Culicidae) in Nagasaki, Japan. Jpn. J. Sanit. Zool., 46: 131–138.
9) Takagi, M. Tsuda, Y. & Wada, Y. (1995b): Temporal and spatial distribution of released *Aedes albopictus* (Diptera: Culicidae) in Nagasaki, Japan. Jpn. J. Sanit. Zool., 46: (in press).
10) Trpis, M. & Hausermann, W. (1986): Dispersal and other population parameters of *Aedes aegypti* in an African village and their possible significance in epidemiology of vectorborne diseases. Am. J. Trop. Med. Hyg., 35: 1263–1279.