［原著］Comparison of four light-trap methods for collecting mosquitoes in Iriomote Island, Ryukyu Archipelago, Japan

Comparison of four light-trap methods for collecting mosquitoes in Iriomote Island, Ryukyu Archipelago, Japan

Takako Toma[1,2], Yukiko Higa[1,3], Takao Okazawa[4] and Ichiro Miyagi[1,5]

[1] *Environmental Health, School of Health Science, Faculty of Medicine, University of the Ryukyus, Nishihara, Okinawa, 903-0215 Japan*

[2] *Center for Asia-Pacific Island Studies, Universtiy of the Ryukyus, 1 Senbaru, Nishihara, Okinawa, 903-0213 Japan*

[3] *Center for International Collaborative Research, Nagasaki University, Sakamoto 1-12-4, Nagasaki, 852-8523 Japan*

[4] *International Student Center, Kanazawa University, Kakuma-machi, Kanazawa, Ishikawa, 920-1192 Japan*

[5] *Laboratory of Mosquito Systematics of Southeast Asia and South Pacific c/o Ocean Health Corporation, 4-21-11 Iso, Urasoe, Okinawa, 901-2132 Japan*

ABSTRACT

Adult mosquito collections using 4 methods were carried out in a forest and residential areas at the east side of Iriomote Island, Ryukyu Archipelago, for 6-8 nights from 20-28 August, 2003. The total number of mosquitoes collected per half night was 62.7 by BLB light-trap with dry ice, 49.0 by BLB light-trap without dry ice, 25.9 by Bulb light-trap with dry ice, and 6.4 by Bulb light-trap without dry ice in the forest area. There was no statistical difference in the total numbers collected by the 4 methods. Similarly, there was no significant difference in the total number of mosquitoes in the residential areas. As the result of cluster analysis, the species composition of mosquitoes collected were similar among the 4 methods in the forest and among the 3 methods except the Bulb light-trap without dry ice in the residential area. The present results indicate that 3 of the 4 light-trap methods, except the Bulb light-trap without dry ice, are useful for mosquito surveys to determine the abundance and species composition. *Ryukyu Med. J., 26(1,2) 39～45, 2007*

Key words: BLB light-trap, Bulb light-trap, Iriomote Island, mosquito collection method, Ryukyu Archipelago

INTORODUCTION

Certain kinds of light and carbon dioxide are efficient mosquito attractants, and have been used in various traps[1-3]. Recently, the battery operated Bulb light-traps plus dry ice method has gained popularity for the surveillance of vector mosquitoes of West Nile virus in Japan[4-8]. On the other hand, the mains-operated light-traps with ultraviolet light (light-trap) have also been used commonly for the studies of seasonal appearance of vector mosquitoes of Japanese encephalitis virus (the *Culex tritaeniorynchus*), and lymphatic filariasis (the *Cx. pipiens* group mosquitoes) in Japan[9-16]. It had been shown that dry ice-baited CDC traps caught more mosquitoes than light-traps[17]. The differentiation of attractiveness to many kinds of mosquitoes by different light-trap collections is essential in mosquito faunal survey. However, information is available only for limited species like vector mosquitoes, *Cx. tritaeniorhynchus* and *Cx. pipiens* group mosquitoes.

The efficiency of 4 different methods-BLB light-trap with dry ice, BLB light-trap without dry ice, Bulb light-trap with dry ice and Bulb light-trap without dry ice was evaluated by comparing mosquito abundance and species composition in mosquito collections from a forest and residential areas

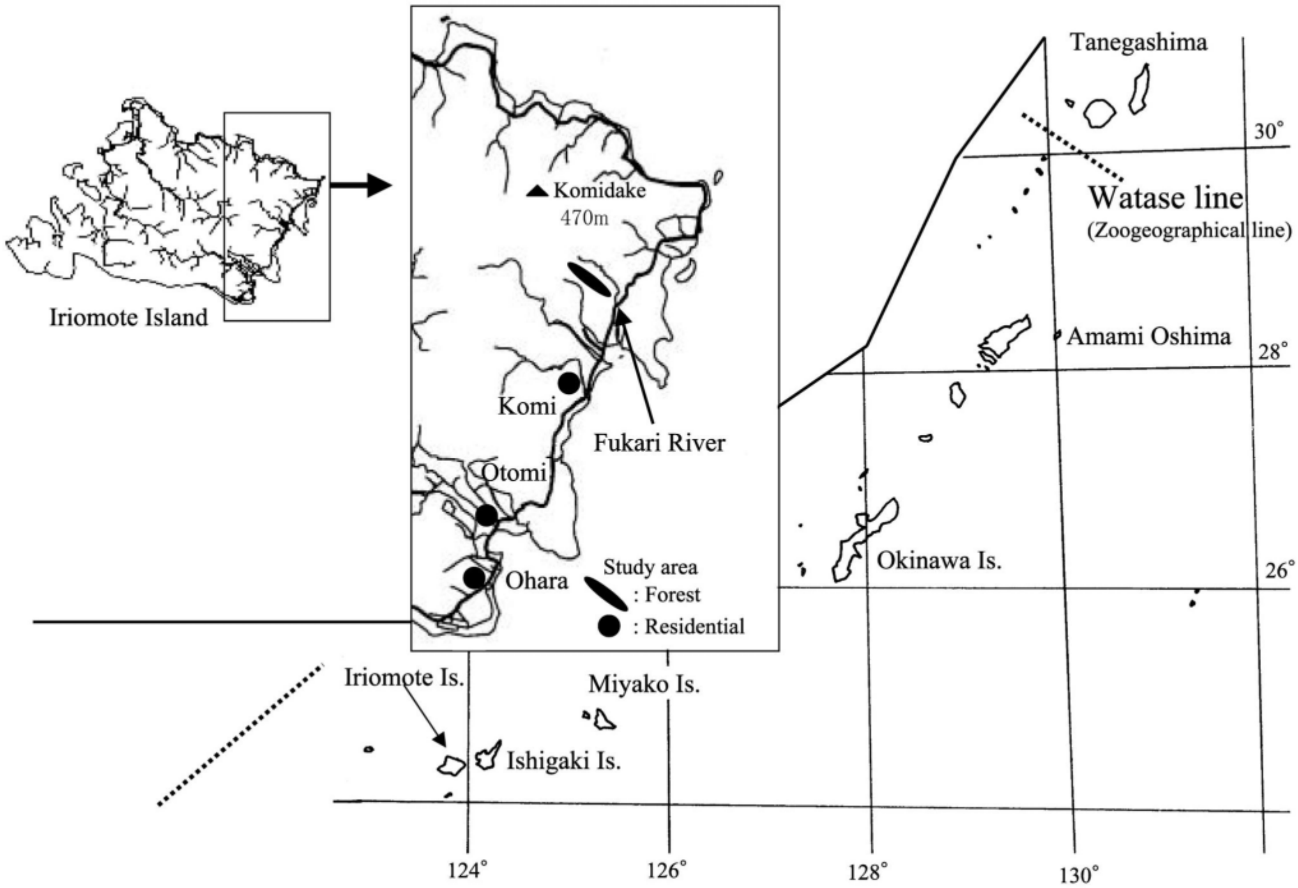


Fig. 1 Location of mosquito collection sites on east side of Iriomote Island, Ryukyu Archipelago, Japan.

of Iriomote Island, Ryukyu Archipelago, Japan, on August 20-28, 2003.

MATERIALS AND METHODS

Iriomote Island (24° 23' N, 123° 45' E), measuring 284.44 km^2, is located approximately 700 km south of Okinawa Island, with about 90 % covered with thick forests (Fig. 1). There are many mangrove trees in the peripheral areas of the island. The climate is subtropical, having an average annual temperature of 23.9℃ (the coldest 17.2 ℃ in February, and hottest 29.5℃ in July) and annual rainfall of 1,329 mm according to the Meteorological Observatory of Okinawa in 2003. The total population is 2,242 according to the census of 2004 by Taketomi Town of Okinawa Prefecture.

Mosquito collections were made for 8 nights from August 20 to 28, 2003 at the east side on Iriomote Island. One study location was selected at a forest area along the Fukari River in Komi Village where there was a small area of paddy field. Other locations were selected at residential areas of villages, namely Ohara, Otomi and Komi.

Two kinds of light-trap (Fig. 2)-(1) *Ishizakidenki-seisakusho*, Japan, a main- or generator-operated black light blue light-trap (abbreviated as BLB light-trap); and (2) *Inokuchi-tekkou*, Japan, with similar design to the CDC battery operated miniature light-trap (abbreviated as Bulb light-trap) - were used for adult mosquito collections[3,4]. The BLB light-trap was a suction trap, with a 30W fluorescent BLB lamp, and was used with or without 1 kg of dry ice. It was operated by a handy generator in the forest area and by electricity in the residential area. The dry ice, wrapped with paper in a Styrofoam-box, was hung close to the light-trap. Carbon dioxide (CO_2) gas diffused continuously through a small gap of the box that preserved the dry ice. The Bulb light-trap, also a suction trap, was used with or without 1 kg of dry ice, operated by batteries. This light-trap consisted of a 3.0-V motor with a three-bladed fan and a 0.3-A light bulb operated by 4 1.5-V dry batteries. The design was shown in

Table 1 Mean number of mosquitoes collected by 2 types of light-traps with or without dry ice in 2 different areas of Iriomote Island, Ryukyu Archipelago, Japan, in August, 2003

	Collection method				2 way ANOVA (Trap x Dry ice)
	BLB + DI	BLB	Bulb + DI	Bulb	
Forest					
Female	45.0 ± 30.3	32.9 ± 7.4	22.5 ± 16.1	5.4 ± 3.4	F=0.829, P>0.05
Male	17.7 ± 7.7	16.1 ± 12.2	3.4 ± 6.7	1.0 ± 1.2	F=0.482, P>0.05
Total	62.7 ± 32.7	49.0 ± 14.5	25.9 ± 19.6	6.4 ± 3.4	F=1.005, P>0.05
Residential area					
Female	6.4 ± 5.4	2.0 ± 2.7	4.5 ± 4.6	0.1 ± 0.7	F=0.020, P>0.05
Male	3.1 ± 2.3	1.4 ± 1.4	1.5 ± 1.9	0.1 ± 0.3	F=0.092, P>0.05
Total	9.5 ± 7.1	3.4 ± 3.8	6.0 ± 5.7	0.4 ± 0.7	F=0.018, P>0.05

BLB+DI: BLB (black light blue) light-trap with dry ice, BLB: BLB light-trap without dry ice, Bulb+DI: Bulb light-trap with dry ice, Bulb: Bulb light-trap without dry ice.

details by Tsuda *et al*[4]. These traps were hung on branches of trees, about 1.5 m above the ground. All light-traps were operated for 6-8 nights from 18:00 to 24:00 h.

In the forest area, 4 collection sites were selected for 4 methods within 200 m along the Fukari River. To avoid disturbance of the light of each trap, the 4 collection sites were very carefully selected. The collection methods were rotated every day to minimize environmental differences among 4 collection sites. Consequently the same type of light-trap was used in a collection site after 4 times of collection.

In the residential area, 4 traps were also set in 4 sites -2 (MY, OK) in Ohara, 1 (ST) in Otomi, and 1 (MT) in Komi. They were changed successively as in the forest area. The collection sites were situated along the sea coast of the east side of the island (Fig. 1). Ohara is a center and the entry point of the east side of the island, and has a large population of 315 inhabitants. Otomi lies halfway between Ohara and Komi and has a population of 292. Komi is 13 km from Ohara, close (2 km) to the forest and having a small population of 66 inhabitants.

Mosquitoes captured by the traps were transported to the field laboratory, Iriomote Wildlife Center, and species identification was performed by methods described by Tanaka *et al*[18], Toma and Miyagi[19] and Miyagi *et al*[20]. The number of mosquitoes and the species collected were compared among the 4 collection methods conducted in/between forest and residential areas. Recently the generic and subgeneric status of Aedine mosquito species were changed by Reinert and Harbach[21], but the names used in the past except some genera and subgenera are adopted to avoid confusion.

RESULTS

The total number of mosquitoes collected for one night was 62.7 by BLB light-trap with dry ice, 49.0 by BLB light-trap without dry ice, 25.9 by Bulb light-trap with dry ice, and 6.4 by Bulb light-trap without dry ice methods in the forest area (Table 1). The numbers of male and female mosquitoes collected by the 4 collection methods were not significantly different. In the residential area, the total number of mosquitoes collected was 9.5 by BLB light-trap with dry ice, 3.4 by BLB light-trap without dry ice, 6.0 by Bulb light-trap with dry ice, and 0.4 by Bulb light-trap without dry ice. There is no significant difference in the numbers of mosquitoes collected by the 4 collection methods.

A total of 1,026 mosquitoes in 25 species was collected in the forest area, and 154 mosquitoes in 17 species in the residential areas (Tables 2, 3). The most common species in the forest were *Uranotaenia macfarlanei* Edwards, *Verrallina iriomotensis* Tanaka & Mizusawa, *Cx. fuscocephala* Theobald and the *Cx. vishnui* subgroup (including *Cx. tritaeniorhynchus* Giles and *Cx. pseudovishnui* Colless), which together comprised 82.3 % of the total collection by the 4 methods. The number of species collected was 18 by BLB light-trap with dry ice, 19 by BLB light-trap without dry ice, 17 by Bulb light-trap with dry ice, and 13 by Bulb light-trap without dry ice. In the residential area, the predominant species were *Aedes albopictus* (Skuse)

Table 2 The number of mosquitoes and species collected by 4 methods in the forest area, Iriomote Island

	BLB+DI				BLB				Bulb+DI				BLB				Grand	%
	female	male	total	%	female	male	total	%	female	male	total	%	female	male	total	%	total	
Anophelessinensis	1	1	2	0.5	0	0	0	0	0	0	0	0	0	0	0	0	2	0.2
An. minimus	2	4	6	1.6	5	1	6	1.5	2	0	2	1.0	0	0	0	0	14	1.4
Mimomyialuzonensis	0	0	0	0	3	1	4	1.0	1	0	1	0.5	1	0	1	2.0	6	0.6
Coquillettidiacrassipes	0	6	6	1.6	0	1	1	0.3	0	0	0	0	0	0	0	0	7	0.7
Uranotaeniaohamai	2	4	6	1.6	0	2	2	0.5	1	0	1	0.5	1	0	1	2.0	10	1.0
Ur. yaeyamana	0	1	1	0.3	2	1	3	0.8	1	0	1	0.5	0	1	1	2.0	6	0.6
Ur. annandalei	0	0	0	0	0	0	0	0	1	0	1	0.5	2	0	2	3.9	3	0.3
Ur. macfarlanei	70	16	86	22.9	146	23	169	43.1	25	0	25	12.1	18	1	19	37.3	299	29.1
Ur. lateralis	3	0	3	0.8	8	0	8	2.0	7	10	17	8.2	1	1	2	3.9	30	2.9
Orthopodomyiaanopheloides	0	1	1	0.3	0	0	0	0	2	0	2	1.0	0	0	0	0	3	0.3
Ochlerotatusa. okinawanus	0	0	0	0	0	1	1	0.3	0	0	0	0	0	0	0	0	1	0.1
Oc. baisasi	2	1	3	0.8	1	1	2	0.5	0	0	0	0	0	0	0	0	5	0.5
Verrallinaatriisimilis	1	0	1	0.3	1	0	1	0.3	2	3	5	2.4	1	1	2	3.9	9	0.9
Ve. iriomotensis	62	3	65	17.3	27	2	29	7.4	94	1	95	45.9	6	0	6	11.8	195	19.0
Ae. riversi	8	2	10	2.7	1	3	4	1.0	8	1	9	4.3	0	0	0	0	23	2.2
Ae. vexansnipponii	2	6	8	2.1	0	2	2	0.5	2	0	2	1.0	0	0	0	0	12	1.2
Armigeressubalbatus	1	0	1	0.3	1	0	1	0.3	0	0	0	0	0	0	0	0	2	0.2
Culexh. ryukyuanaus	0	0	0	0	0	1	1	0.3	0	0	0	0	0	0	0	0	1	0.1
Cx. cinctellus	0	13	13	3.5	0	21	21	5.4	0	1	1	0.5	0	1	1	2.0	36	3.5
Cx. infantulus	0	0	0	0	0	2	2	0.5	0	2	2	1.0	0	1	1	2.0	5	0.5
Cx. fuscocephala	50	40	90	23.9	30	33	63	16.1	10	6	16	7.7	11	2	13	25.5	182	17.7
Cx. quinquefasciatus	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Cx. bitaeniorhynchus	0	1	1	0.3	0	0	0	0	3	1	4	1.9	1	0	1	2.0	6	0.6
Cx. vishnui subgroup*	66	7	73	19.4	38	34	72	18.4	21	2	23	11.1	1	0	1	2.0	169	16.5
Total	270	106	376	100	263	129	392	100	180	27	207	100	43	8	51	100	1026	100
No.ofspeciescollected			18				19				17				13		24	

BLB+DI: BLB(blacklightblue) light-trap with dry ice, BLB-DI: BLB light-trap without dry ice, Bulb+DI: Bulb light-trap with dry ice, Bulb-DI: Bulb light-trap without dry ice. BLB light-trap with dry ice was carried out 6 times, and other methods were carried out 8 times.

**Culex vishuni* subgroup includes *Cx. tritaeniorhynchus* and *Cx. psuedovishnui.*

Table 3 The number of mosquitoes and species collected by 4 methods in the residential area, Iriomote Island

	BLB+DI				BLB				Bulb+DI				Bulb				Grand	%
	female	male	total	%	female	male	total	%	female	male	total	%	female	male	total	%	total	
An. sinensis	1	0	1	1.3	0	0	0	0	0	0	0	0	0	0	0	0	1	0.6
An. minimus	2	1	3	3.9	0	0	0	0	0	0	0	0	0	0	0	0	3	1.9
Mi. luzonensis	2	0	2	2.6	0	0	0	0	0	0	0	0	0	0	0	0	2	1.3
Cq. crassipes	1	0	1	1.3	0	2	2	7.1	0	0	0	0	0	0	0	0	3	1.9
Ur. annandalei	0	0	0	0	0	0	0	0	0	0	0	0	1	0	1	50.0	1	0.6
Ur. macfarlanei	0	1	1	1.3	0	0	0	0	0	0	0	0	0	0	0	0	1	0.6
Ur. lateralis	1	0	1	1.3	0	0	0	0	0	0	0	0	0	0	0	0	1	0.6
Oc. baisasi	2	3	5	6.6	3	0	3	10.7	0	0	0	0	0	0	0	0	8	5.2
Ae. albopictus	11	7	18	23.7	6	4	10	36.7	17	8	25	52.1	0	0	0	0	53	34.4
Ae. f. miyarai	0	0	0	0	0	1	1	3.6	0	0	0	0	0	0	0	0	1	0.6
Ae. riversi	1	2	3	3.9	0	1	1	3.6	3	1	4	8.3	0	0	0	0	8	5.2
Ae. vexans nipponii	0	0	0	0	1	0	1	3.6	0	0	0	0	0	0	0	0	1	0.6
Ar. subalbatus	21	5	26	34.2	4	1	5	17.9	13	3	16	33.3	0	0	0	0	47	30.5
Cx. fuscocephala	2	2	4	5.3	1	0	1	3.6	0	0	0	0	0	0	0	0	5	3.2
Cx. quinquefasciatus	1	4	5	6.6	0	2	1	7.1	1	0	1	2.1	1	0	1	50.0	9	5.8
Cx. vishnui subgroup*	6	0	6	7.9	1	1	1	7.1	2	0	2	4.2	0	0	0	0	10	6.5
Total	51	25	76	100	16	12	28	100	36	12	48	100	2	0	2	100	154	100
No. of species collected			13				10				5				2		16	

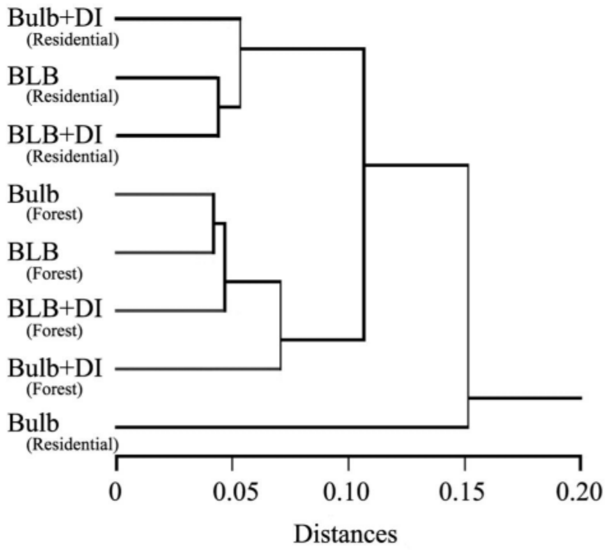
BLB+DI: BLB (black light blue) light-trap with dry ice, BLB: BLB light-trap without dry ice, Bulb+DI: Bulb light-trap with dry ice, Bulb: Bulb light-trap without dry ice. Every method was carried out 8 times.

**Culex vishuni* subgroup includes *Cx. tritaeniorhynchus* and *Cx. psuedovishnui.*

and *Armigeres subalbatus* (Coquillett), comprising 64.9 % of the total collection. The number of species collected was 13 by BLB light-trap with dry ice, 10 by BLB light-trap, 5 by Bulb light-trap with dry ice, and 2 by Bulb light-trap. The number of species collected by Bulb light-trap without dry ice was small in both areas.

By cluster analysis, the species composition of mosquitoes collected by the 4 methods was similar within the forest area, but different from those in

Fig. 2 BLB light-trap (left) and Bulb light-trap (right) used in the survey.



BLB+DI: BLB (black light blue) light-trap with dry ice, BLB: BLB light-trap without dry ice, Bulb+DI: Bulb light-trap with dry ice, Bulb: Bulb light-trap without dry ice.

Fig. 3 Results of cluster analysis to species composition of mosquitoes collected by 4 light-trap methods in forest and residential areas in Iriomote Island.

the residential area. Within the residential area, the species composition was similar by 3 methods except Bulb light-trap without dry ice. The trees obtained from the results were similar in 5 except Centroid of 6 methods (Single, Complete, Centroid, Average, Median and Ward) - see as example the Single linkage method (Fig. 3).

DISCUSSION

Light-traps for adult mosquito collection have been used for many years to catch insects, and numerous papers have been published dealing with various trap designs, fans, motors, different light source characteristics, etc[3]. In Japan, these are the mains-operated ultraviolet light-traps that have been used both inside and outside animal shelters for collecting vectors in Japanese encephalitis endemic areas. These traps have proved successful in catching large numbers of *Cx. tritaeniorhynchus* and other mosquitoes such as *Anopheles sinensis* Widemann, *Ae. vexans nipponii* (Theobald) and *Cx. pipiens pallens* Coquillett[9-15].

It is now generally accepted that carbon dioxide is an attractant and since mid 1970s, there has been increasing usage of carbon dioxide for mosquito collection[3]. With the introduction of battery operated CDC miniature light-traps and the availability of dry ice, there has been renewed interest in combining light with carbon dioxide in mosquito traps. This light-traps supplemented with carbon dioxide have widely been used recently for surveillance programmes of the West Nile virus in Japan[6-8].

There appear to be no reports regarding comparison of mosquito number and species by the 4 methods, BLB light-trap with or without dry ice, and Bulb light-trap with or without dry ice in Japan. In the present survey, a total of 1,026 with 25 species was collected in the forest area of Komi for 6-8 nights in August by 4 different light-traps. *Uranotaenia macfarlanei* whose larvae breed in streams, *Ve. iriomotensis* breeding in ground pools in the forest, and *Cx. fuscocephala* and *Cx. vishnui* subgroup breeding in paddy fields, were collected abundantly in all 4 methods in the forest area. In 1978, mosquito collections carried out at the same area in the forest of Komi Village, for 9 nights in July and October, by using only BLB light-trap without dry ice yielded a total of 1,200 mosquitoes of 23 species[22]. The most common species in the survey of 1978 were *An. sinensis* (44.5%), *Cx. tritaeniorhynchus* (25%) and *Cx. cinctellus* Edwards (11.3%). *Ur. macfarlanei*, and these species were not collected in the present study. At that time, there was a vast expanse of paddy fields in the forest area compared with the present study, and a lot of paddy field breeders were collected.

On the other hand, the mosquito fauna in the residential area with 17 species was poor compared to the forest. Among them, *Ae. albopictus* and *Ar. subalbatus* were commonly collected. The number of *Cx. quinquefasciatus* Say that had been a common species in residential area, Naha, Okinawa, was small[16]. In the residential area of Iriomote Island, the forest species like *An. minimus* Theobald (whose larvae breed in the streams run out in the peripheral areas of the forest), *Ae. riversi* Bohart & Ingram (a tree hole breeder in the forest), *Ur. ohamai* Tanaka *et al* (a fresh water crab hole breeder in the forest and brackish water crab hole breeder in mangrove forest), and *Ochlreotatus baisasi* Knight & Hull (a brackish crab hole breeder in mangrove forest) were also collected[23].

In the present survey, although the BLB light-trap with dry ice tended to increase the catch of mosquitoes, there was no statistical difference in the numbers collected by the 4 collection methods.

Magnarelli (1975) mentioned that the best trap would probably be a combination of light and dry ice[17]. Apparently, BLB mains-operated light-traps catch a large number of many kinds of small insects including mosquitoes. Mosquitoes get damaged when they pass through the fan of the trap and as a result, sorting mosquitoes in a large number of insects and species identification are difficult and time consuming. Moreover, electricity and dry ice are often not available in remote study areas. On the other hand, the battery operated Bulb light-trap is small and can easily be carried around in remote study areas. However the present survey revealed that the Bulb light-trap was not effective as reflected by the species composition. Very recently, Saito and his colleagues have developed a new convenient method to supply carbon dioxide for mosquito sampling by using yeast which converts sugar into carbon dioxide and ethyl alcohol[24]. To compensate for the weak point of the Bulb light-trap, yeast-generated carbon dioxide trap might be valuable for mosquito faunal study in remote areas. Although the number of species collected by the Bulb light-trap without dry ice was small in the forest and residential areas, the other 3 methods, namely BLB with or without dry ice and the Bulb with dry ice of the 4 light-trap methods tested in the present study were similar for the number of mosquitoes and proved useful for collecting various species belonging to the genera *Culex, Aedes, Ochlreotatus* and *Uranotaenia*. These 3 methods could be useful for mosquito survey to determine the mosquito number, fauna and species composition. We could choose any of these 3 mosquito collection methods depending on the condition of electricity supply, availability of dry ice, access to the survey area, and other factors.

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