

SURVEILLANCE AND BEHAVIORAL INVESTIGATIONS OF *Aedes aegypti* AND *Aedes polynesiensis* IN MOOREA, FRENCH POLYNESIA, USING A STICKY OVITRAP

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ABSTRACT. The effectiveness of the sticky ovitrap was assessed for the container-breeding *Aedes aegypti* and *Aedes polynesiensis* in Moorea, French Polynesia. These mosquitoes are the primary vectors of dengue viruses and Bancroftian filariasis, respectively, in the area. Both *Ae. aegypti* and *Ae. polynesiensis* were collected in greatest numbers in sticky ovitraps baited with water or grass infusions rather than leaf infusions. Sticky ovitrap collections were significantly higher for both species in the 12 h post-midday than pre-midday and in traps set in shaded compared with open locations. More females of *Ae. aegypti* were collected in ovitraps at west-facing walls, although *Ae. polynesiensis* collected at east- or west-facing traps did not differ in number. Female *Ae. aegypti* (bloodfed, marked, and released for oviposition) were readily recaptured (19–26%) by sticky ovitraps, exhibiting movement of up to 30 m, and between outdoor and indoor situations. Overall, the sticky ovitrap proved an effective tool for investigating the oviposition behavior and dispersal of these container-breeding species.

KEY WORDS *Aedes aegypti*, *Aedes polynesiensis*, oviposition, dispersal, sticky ovitrap

INTRODUCTION

The sticky ovitrap (Ritchie et al. 2003) has been shown to be as sensitive as a standard ovitrap in detecting the presence of *Aedes aegypti* (L.) at domestic premises, is effective both inside and outside buildings, and allows ready identification of captured specimens. Ritchie et al. (2003, 2004) suggested that, apart from their use as a lethal surveillance tool, the traps would be useful for ecological and epidemiological investigations. To that end, the effectiveness of the sticky ovitrap for surveillance and behavioral study of container-breeding species of concern as vectors of disease in the Pacific Islands region was investigated in Moorea, French Polynesia. *Aedes aegypti* and *Aedes polynesiensis* Marks, the former being the principal vector of dengue viruses in the region and the latter a vector of *Wuchereria bancrofti* and a secondary vector of dengue viruses, were both present on the island.

Aedes aegypti in Moorea is primarily associated with artificial container habitats in domestic residential or other urban situations (e.g., drums, tins), although larvae are also found in some peridomestic habitats such as discarded tires. *Aedes polynesiensis* is primarily associated with natural containers in shoreline habitat (e.g., crab holes) and native

forest habitat (e.g., coconut shells and husks, tree holes, palm fronds), but larvae are also found in artificial containers in domestic and peridomestic habitats (e.g., drums and tins, tires, and canoes). The relative influence of attractant infusions within the ovitrap was assessed, and spatial and temporal aspects of local oviposition behavior of both species were investigated. A mark–release–recapture experiment was conducted to examine the dispersal of *Ae. aegypti* in Moorea.

MATERIALS AND METHODS

The sticky ovitraps (U.S. patent pending) were as described previously (Ritchie et al. 2003): a 1.2-liter plastic “golf divot bucket,” although in this study they were of a dark red color rather than black. The adhesive plastic strips were approximately 50 cm in width and attached to the inside of the bucket with clothes pegs; water was added to fill the bucket to the base of the strip.

Study site: The ovitraps were deployed in various sites at the University of California (Berkeley) Gump Station, Pao Pao, Moorea, where human bait collections showed *Ae. aegypti* and *Ae. polynesiensis* actively biting. The sites were adjacent to residential and associated buildings and within 100 m of the Cooks Bay shoreline and native forest habitat (typically hibiscus and coconut).

Site 1 (the workshop clearing) was adjacent to residential and associated buildings and within 50 m of shoreline and native forest habitat. Site 2 (the botanical garden) was adjacent to shoreline and native forest habitat, with housing and associated buildings 100 m distant, although some artificial containers were found within the site. Site 3 (the hillside compound) comprised the Director’s residence and additional residential and other buildings, overlooked sites 1 and 2, and was surrounded by coastal forest. Site 1 had greater habitat oppor-

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tunity and relatively more *Ae. aegypti* activity, whereas site 2 had relatively greater *Ae. polynesiensis* habitat and activity. Site 3 had little habitat opportunity for *Ae. aegypti*, but *Ae. polynesiensis* was common.

Trials were conducted to examine the effects of several factors on collection of female *Ae. aegypti* and *Ae. polynesiensis* with sticky ovitraps.

Effect of infusions: The effectiveness of ovitraps is reportedly enhanced by the use of a vegetation-based infusion (Ritchie 2001, Ritchie et al. 2003), so water quality preferences for oviposition were investigated at sites 1 and 2. The traps were assessed by the triangular placement of 3 traps 3 m apart within each site, with either clean household tap water (unchlorinated) left standing outdoors for at least 3 days in 5-liter plastic uncapped containers or the same infused with approximately 500 cc of a rough-cut loose-pack of either local fresh green grass (T grass, *Paspalum conjugatum*) or dead leaf of the two locally common broadleaf trees (mango, *Mangifera indica*, and hibiscus, *Hibiscus tiliaceus*) for 7 days. The traps were left out for 9 days, with the position of each trap (with its contents) being rotated every 3 days to compensate for position effects. The trial was undertaken in September 2003 and repeated in October 2003.

Timing of oviposition: The ovitraps were used also to investigate temporal and spatial oviposition activity at sites 1 and 2. A mixture of clean water and grass- and leaf-infused water (2:2:1) was used in all ovitraps, a decision made to accommodate both species in accordance with the 1st month's results in the initial investigation. Four ovitraps each were placed randomly in wind-sheltered situations at sites 1 and 2 and were collected and replaced at midday and midnight over 5 days in November 2003 to investigate the relative timing of oviposition activity.

Trap exposure (sunlit and open or shaded and sheltered): Four ovitraps each were placed randomly in 2 wind-sheltered situations at site 2, either among the roots of banyan trees (*Ficus benghalensis*) or in open space with no overhead tree canopy, and collected after 5 days in September 2003, with the exercise repeated in October 2003.

Trap orientation (east or west trap exposure): Three traps in a line and 2 m apart were placed in wind-sheltered sites in front of east- and west-facing walls of buildings near site 1 to investigate solar influences on oviposition. The traps were left for 5 days in September 2003, and the exercise was repeated in October 2003.

Analysis: The data from the trials for effect of infusion and timing of oviposition were analyzed by 2-way ANOVA for site, treatment, and their interactions. Tukey's multiple comparison procedure was used to separate means. An unpaired *t*-test was used to compare means in the trap exposure and orientation trials. The data were $\log(X + 1)$ transformed if not normally distributed. The data from

the temporal and spatial trials were tested for significance with an unpaired *t*-test.

Mark-release-recapture study of *Ae. aegypti* dispersal: Dispersal for oviposition was investigated in November 2003 with a mark-release-recapture exercise at site 3 after the area had been cleared (by removal over 2 weeks of all actual and potential artificial containers suitable for *Ae. aegypti* oviposition and larval development) and human bait collections had not revealed adults of the species. Larvae of *Ae. aegypti*, collected from domestic containers or hatched from eggs of wild adults, were reared to adults that were held on sugar solution for 2 days to allow for mating and were then fed blood. The bloodfed adults were split into 2 cohorts of approximately 100 females in screened plastic containers, and each cohort was dusted with a different colored fluorescent powder (Radglo green RS11 and Radglo magenta RS 18) with a modified food baster. The following day, 1 cohort was released at an internal location and the other at an external location within the residential compound after 20 ovitraps had been placed strategically (between and within the buildings) around the compound up to 80 m from the release sites (Fig. 1). The ovitraps were collected after 5 days and examined for adult *Ae. aegypti* with colored dust that revealed their release point. This investigation could not be undertaken similarly for *Ae. polynesiensis* because the species and its natural habitats (crab-holes, coconut husks and shells, tree holes) were ubiquitous, and no suitable study area free of the species and without alternative (to the ovitraps) oviposition habitats could be found in the locality.

RESULTS

Effect of infusions: The collection of gravid *Ae. aegypti* and *Ae. polynesiensis* was significantly ($P < 0.05$) affected by infusion type, with leaf infusions least preferred by both species (Table 1).

Timing of oviposition: A significantly larger number of each species was collected in traps set in the post-midday time sectors ($P < 0.05$; Table 1).

Trap exposure: Shaded, sheltered sticky ovitraps collected significantly ($P < 0.05$) more female *Ae. aegypti* and *Ae. polynesiensis* than did ovitraps in sunlit open areas (Table 1).

East or west trap exposure: More female *Ae. aegypti* were collected in traps at west-facing rather than east-facing walls ($P < 0.05$), but numbers of *Ae. polynesiensis* collected at east- or west-facing traps were not different (Table 1).

Mark-release-recapture study of *Ae. aegypti* dispersal: The recapture rate of marked *Ae. aegypti* was 19% and 26% for the external and internal releases, respectively. Dispersal was relatively limited in the external environment, with the furthest collections being no more than 30 m distant by shortest measure from the release point. Some

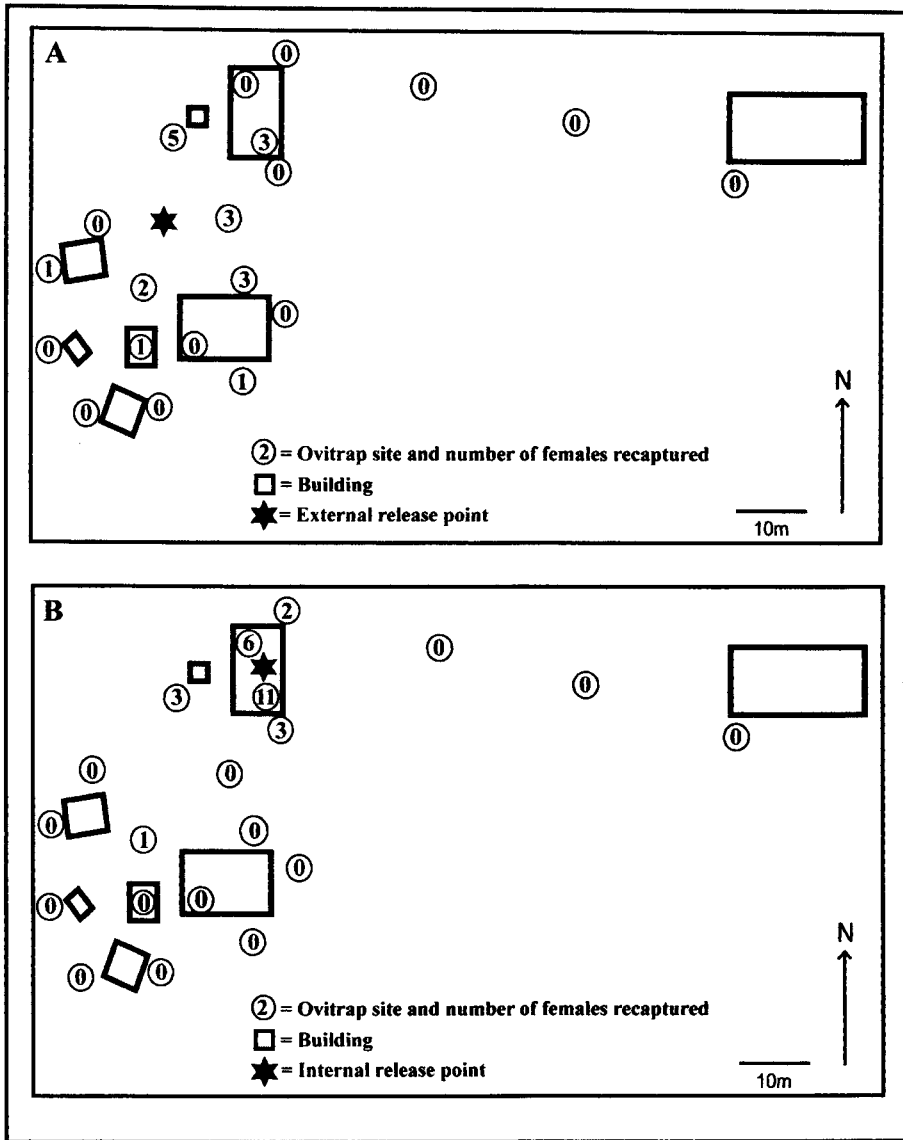


Fig. 1. Maps of residential compound with location of external (A) and internal (B) release points for marked and bloodfed *Aedes aegypti*, and the locations of ovttraps and numbers of females recaptured therein during November 2003 at Gump Station, Pao Pao, Moorea, French Polynesia.

movement occurred from the external release site into the buildings, with 4 (21%) of the 19 recaptures in internal ovttraps being from the external release site (Fig. 1a). Movement was relatively greater from the internal release site to the outside environment, with 9 (35%) of the 26 recaptures in external ovttraps being from the internal release, although all but one were taken relatively close to the release point (Fig. 1b).

DISCUSSION

Aedes aegypti has been reported as being more attracted to containers (and ovttraps) when the wa-

ter contained some organic matter (e.g., Bond and Fay 1969, Reiter and Gubler 1997, Ritchie 2001), but in this investigation, the species seemingly did not prefer the grass infusion to clean water. The preference showed by *Ae. polynesiensis* for grass over leaf infusion was similar to that reported by Gubler (1971), who, however, also found a preference for dark color. The leaf infusion in this study was dark colored but was also strong smelling, whereas the grass infusion had only a slight odor and was not greatly dissimilar to the clean water in clarity. It is possible the leaf infusion was too concentrated with ammonia and protein, which was found by Gubler (1971) to be repellent to ovipos-

Table 1. Summary of experiments with sticky ovitraps from September to October 2003 at Gump Station, Pao Pao, Moorea, Tahiti. Trials were conducted at 2 sites, and 2-way ANOVA was used to examine the effect of treatment and site for female *Aedes aegypti* and *Aedes polynesiensis*, except in the trap exposure and orientation trials, in which only 1 site was used and an unpaired *t*-test was employed to compare means.¹

Experiment (traps/treatment)	<i>Ae. aegypti</i>		<i>Ae. polynesiensis</i>	
	Mean \pm SE	<i>P</i> values	Mean \pm SE	<i>P</i> values
Infusion type (12)				
Plain water	3.75 \pm 0.69 a	Treatment: 0.007	4.58 \pm 0.68 ab	Treatment: 0.012
Grass	3.08 \pm 0.56 ab	Site: <0.001	5.75 \pm 0.86 a	Site: 0.058
Leaf	2.00 \pm 0.35 b	Treat \times site: 0.869	2.67 \pm 0.51 b	Treat \times site: 0.720
Time of oviposition (8)				
Pre-midday	0.38 \pm 0.18 a	Treatment: <0.001	1.63 \pm 0.26 a	Treatment: <0.001
Post-midday	2.87 \pm 0.55 b	Site: 0.074	4.38 \pm 0.65 b	Site: 0.025
		Treat \times site: 0.167		Treat \times site: 0.225
Trap exposure (8)				
Trap in shady site	2.88 \pm 0.30 a	<i>t</i> = 7.201	5.88 \pm 0.77 a	<i>t</i> = 5.945
Trap in exposed site	0.38 \pm 0.18 b	<i>P</i> < 0.001	1.13 \pm 0.23 b	<i>P</i> < 0.001
Trap orientation (12)				
Trap east facing	2.50 \pm 0.44 a	<i>t</i> = 2.351	3.25 \pm 0.43 a	<i>t</i> = 0.343
Trap west facing	3.92 \pm 0.42 b	<i>P</i> = 0.028	3.00 \pm 0.59 a	<i>P</i> = 0.735

¹ Numbers followed by a different letter indicates means are significantly different (*P* < 0.05) by Tukey's multiple comparison test or *t*-test.

iting *Ae. polynesiensis* and *Ae. albopictus*. This interpretation might be supported by the fact that no males were found at the leaf infusion traps, although they were collected occasionally at both other waters.

The sticky ovitrap proved useful for investigating the oviposition behavior of both *Ae. aegypti* and *Ae. polynesiensis*. The pre-midday and post-midday collections from this investigation enhance previous knowledge of the activity of the 2 species. For *Ae. polynesiensis*, no data on oviposition activity had been published previously. For *Ae. aegypti*, oviposition in the laboratory has been reported during the morning, although peak activity occurred in the afternoon, with the time for egg laying determined by the effect of light (Haddow and Gillett 1957). From the field, there have been reports of less oviposition by *Ae. aegypti* in the 2 hours after sunrise than in that period before sunset in Trinidad (Chadee and Corbet 1987, Corbet and Chadee 1990).

Both species were collected by the ovitraps in sheltered and shaded positions, supporting the findings of McKenzie (1925) in Rarotonga and Symes (1960) in Fiji that *Ae. polynesiensis* adults prefer sheltered areas. For *Ae. aegypti* in the Pacific region, partially shaded containers were reported as being ideal breeding places in Hawaii (Bohart and Ingram 1946, cited in Lee et al. 1987). The westerly facing traps collected more *Ae. aegypti*, but *Ae. polynesiensis* appeared to be nonselective in this trial for any solar influence in oviposition. Corbet and Chadee (1990) have reported more ovipositioning by *Ae. aegypti* in sites facing west compared with east in Trinidad, although an earlier report (Evans and Bevier 1969) found no such preference for ovi-

traps facing north, south, east, or west at 3 locations in the southern United States.

The mark-release-recapture investigation was intended to "track" gravid females seeking an ovipositing site following their release. However, *Ae. aegypti* females are known to feed more than once in each gonotrophic cycle (Macdonald 1956, Scott et al. 1993), and some host-seeking activity could have occurred prior to their capture at the ovitrap, precluding a simple interpretation of the results. At night, no humans were in residence in the main cluster of buildings surrounding the release points at the time of the release, although people worked in the buildings during the daytime, 2 dogs were present intermittently, and one of us (RCR) was resident in the house in the northeast of the compound (although no marked adults were collected by the ovitraps near this building). It is possible that if the ovitraps had been left out longer, more marked adults might have been collected and at more distantly sited ovitraps. The recapture rates recorded here (19% of the external and 26% of the internal releases) fall within the general range of others reported in the literature (12.4%, Conway et al. 1974; 37.5%, McDonald 1977; 5.4%, Nayar 1981; 40%, Trpis and Hauserman 1986), although these investigations used different techniques. In a recent and more similar mark-release-recapture study over 7 days, sticky lures recaptured 2.7% and 8.7% of females released outdoors and indoors, respectively (Muir and Kay 1998), but those results also are not strictly comparable to the present study because those females were not bloodfed when released and the sticky lures were collecting adults seeking resting, not ovipositing, sites.

The females recaptured were in ovitraps situated no further than 30 m from the release points. Indeed, a majority (65%) of the recaptured internally released mosquitoes was collected within the release building. These might have included females initially seeking a resting site rather than an ovipositing site, although the building had numerous suitable resting sites. The mosquitoes might have dispersed further under different conditions or when looking for a bloodmeal rather than an ovipositing site. Distance traveled for oviposition will be dependent on the duration of the gonotrophic cycle and the availability of ovipositing sites (Edman et al. 1998). The duration of the *Ae. aegypti* gonotrophic cycle has been reported to vary from <2 days to >5 days (Macdonald 1956), and generally, *Ae. aegypti* has been reported to disperse not far if its resources for feeding and ovipositing are available nearby (Teesdale 1955). Sheppard et al. (1969) estimated that *Ae. aegypti* females moved an average of 37 m in 24 h in Bangkok; Nayar (1981) proposed dispersal of <100 m in Florida; Muir and Kay (1998) reported mean distance traveled per day over 7 days was 16.8 m and 24.7 m for indoor and outdoor releases, respectively, in Australia; Harrington et al. (2001) estimated a range of 79 m in Puerto Rico and Thailand; and Getis et al. (2003) found that the species clustered within houses and out to ~30 m in Peru. However, Reiter et al. (1995) reported much greater dispersal in Puerto Rico and suggested that individual females dispersed, on average, 279 m after 5 days and covered an area of 840 m diameter while searching for blood and ovipositing sites. The relatively short distances reported here for Moorea are more consistent with the other studies but do not necessarily indicate the typical range of dispersal for *Ae. aegypti* in French Polynesian communities. Further studies are required to elucidate this vector risk to local populations.

Overall, the sticky ovitrap proved effective in collecting both *Ae. aegypti* and *Ae. polynesiensis* on Moorea and proved to be useful for investigating ovipositing behavior of the 2 *Aedes* vector species and their dispersal. The particular advantage of the sticky ovitrap for surveillance in situations such as Moorea would be not in the quantitative monitoring of populations of common species such as *Ae. polynesiensis*, but for the detection and monitoring of *Ae. aegypti* (and perhaps *Ae. albopictus*, which has displayed similar ovipositing behavior to *Ae. polynesiensis* [Gubler 1971] and has been collected readily with sticky ovitraps in Vietnam [Ritchie, unpublished data]) when it is at low levels following source reduction or other control efforts or when introduction to localities where it does not exist is a risk and requires quarantine border control programs.

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