

FIELD EFFICACY OF THE BG-SENTINEL COMPARED WITH CDC BACKPACK ASPIRATORS AND CO₂-BAITED EVS TRAPS FOR COLLECTION OF ADULT *Aedes aegypti* IN CAIRNS, QUEENSLAND, AUSTRALIA

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ABSTRACT. In this study, we compared the efficacy of the newly available BG-Sentinel with an established “gold standard,” the CDC Backpack Aspirator, and a CO₂-baited EVS trap for the collection of *Aedes aegypti* (L.) in Cairns, Australia. BG-Sentinels collected significantly more ($P = 0.017$) female *Ae. aegypti* (mean per collection, 1.92 ± 0.39) than both the CDC Backpack Aspirator (1.00 ± 0.35) and the EVS trap (0.71 ± 0.27). Male-only and combined male-female *Ae. aegypti* collections for the BG-Sentinel and the CDC Backpack Aspirator were also greater than EVS trap collections. The CDC Backpack Aspirator and the BG-Sentinel captured proportionally fewer females compared with the EVS trap. The BG-Sentinel was the most *Ae. aegypti*-specific collection method. The CDC Backpack Aspirator collected proportionally more bloodfed *Ae. aegypti* than the other methods, which collected a greater proportion of nullipars. The data presented here will aid researchers in deciding what *Ae. aegypti* sampling device best suits their needs. BG-Sentinels and CDC Backpack Aspirators should be considered as alternatives to human-bait collections for *Ae. aegypti* sampling.

KEY WORDS *Aedes aegypti*, BG-Sentinel, CDC Backpack Aspirator, EVS trap, Australia

INTRODUCTION

The vector of dengue in Australia, *Aedes aegypti* (L.) (Ritchie et al. 2002) is common throughout cities and towns in northern Queensland, including Cairns (population ca. 125,000), where dengue epidemics frequently occur. Vector control officers in northern Queensland conduct *Ae. aegypti* surveillance by yard inspections for containers containing immatures and by using sticky ovitraps for adults (Ritchie et al. 2003). However, yard inspections are labor-intensive, and sticky ovitraps require a 4–7-day deployment but do not typically yield large numbers of *Ae. aegypti*. Thus, there is a need to make *Ae. aegypti* surveillance in northern Queensland more labor-efficient and productive. In addition, there is a need to develop and refine measures of adult dengue vector abundance to make them more efficient, standardized, and epidemiologically relevant (Focks 2003).

Human-bait collections have repeatedly been shown to be more sensitive for *Ae. aegypti* sampling than traps (Canyon and Hii 1997, Jones et al. 2003, Schoeler et al. 2004). However, there are ethical and safety concerns, particularly during dengue outbreaks, that make this method undesirable for *Ae. aegypti* surveillance. Other potential *Ae. aegypti* collection devices and methods must therefore be considered. Of note are the Fay-Prince trap (Fay and Prince 1970) and the carbon dioxide (CO₂)-baited Centers for Disease Control (CDC) light

trap, both of which have been demonstrated to catch *Ae. aegypti* in quantities that warrant consideration. Fay-Prince traps are generally more effective than CDC light traps (Kloter et al. 1983, Jensen et al. 1994). Neither trap works as well without CO₂, which can be expensive, difficult to obtain and dispense, and is therefore undesirable for use in surveillance programs. In addition, neither trap is as effective as the CDC Backpack Aspirator (Clark et al. 1994), which collected a similar amount of *Ae. aegypti* as human-bait landing collections in a comparative study (Schoeler et al. 2004). The CDC Backpack Aspirator permits indoor collection in common *Ae. aegypti* harborage sites, and it has been used to identify household risk factors for dengue infection (Rodriguez-Figueroa et al. 1995) and to quantify *Ae. aegypti* movement (Edman et al. 1998). This device can be considered the “gold standard” sampling tool for adult *Ae. aegypti*.

Despite these positive attributes, the CDC Backpack Aspirator may not always be ideal for *Ae. aegypti* surveillance. Its weight (12 kg) makes it tiresome to use, and its effectiveness varies between operators. Sampling may be compromised by refused access into the house or to particular rooms. In Cairns, a large proportion of residents are not at home during working hours, making access problematic. Because of its size, there is a risk of property damage when using the aspirator indoors and the method is also personally invasive, because collections typically last 10–30 min (Clark et al. 1994, Schoeler et al. 2004).

An alternative *Ae. aegypti* collection device, the BG-Sentinel (www.bg-sentinel.com) has recently become available. This device does not require carbon dioxide (CO₂) and has shown potential for *Ae. aegypti* surveillance in field trials in Brazil (Kroeckel et al. 2006) and pilot studies in Cairns

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(Williams, unpublished data). However, formal comparison with other methods is required.

We compared the efficacy of the BG-Sentinel and the CDC Backpack Aspirator for the collection of *Ae. aegypti* in Cairns. We also included a CO₂-baited mosquito trap in the comparison. Given that numerous comparisons involving CDC light traps, Fay-Prince traps, and human landing catches have already been made, we selected a CO₂-baited Encephalitis Virus Surveillance (EVS) trap (Rohe and Fall 1979) for comparison. The EVS trap was chosen due to its recent use in *Ae. aegypti* surveillance in northern Australia (Whelan et al. 2005) and French Polynesia (Russell 2004). There also have been no published comparisons of CDC Backpack Aspirators with EVS traps.

MATERIALS AND METHODS

Sampling program

Testing was conducted in the lower level of 2-story "Queenslander"-style houses in Cairns. Such houses are mostly timber, with the well-ventilated lower story typically comprising a laundry, a secondary bedroom, and a garage. The houses used in this study were not fully enclosed by walls and doors on the lower level. Such house design is conducive to *Ae. aegypti* harborage because fly-screened windows and doors are rare, and there are ample refugia and human hosts for mosquitoes in the lower level.

A 4 × 4 Latin Square experimental design (Cochran and Cox 1957) was used, whereby the treatments (CDC Backpack Aspirator, BG-Sentinel, and EVS trap) were rotated among houses to account for positional effects. Two other largely unsuccessful methods shared the 4th treatment position, but they were not included in the analysis because their extremely small *Ae. aegypti* collections may have skewed the data and exaggerated statistical differences. This analysis was repeated six times, resulting in 24 replicates (i.e., 24 houses) for each treatment. A minimum of 2 days separated collections at a house, preventing possible "trap down" effects. The experiment was conducted from March to August 2005.

Sampling devices

The CDC Backpack Aspirator (JW Hock, Gainesville, FL) was used with a 2-person operating team as described by Clark et al. (1994). In this method, the aspirator operator proceeded first, sampling typical *Ae. aegypti* harborage sites such as under, inside, and behind furniture and among clothing, bedding, and any other structures. The second person followed closely with a 23-cm-diameter sweep net to collect any mosquitoes disturbed by the aspirator that had escaped collection. Sampling duration was 10 min, which was suffi-

cient time to ensure complete coverage of the lower level of the house.

The BG-Sentinel (Biogents GmbH, Regensburg, Germany) (Geier et al. 2004, Kroeckel et al. 2006) is a counterflow geometry mosquito collection device that incorporates visual, anemotactic, and olfactory attractants. The BG-Sentinel is a white, cylindrical "bucket" (40 cm in height, 20 cm in diameter) with a white gauze top. In the top center of the trap is a black 10-cm-diameter opening to a catch bag. A 12-V DC fan draws air (and mosquitoes) in through the black opening and exhausts it through the perimeter of white gauze at the top of the trap, creating an air plume that mimics convection currents from the human body (Geier, personal communication). The olfactory attractant, a blend of lactic acid, ammonia, and caproic acid, is placed in the bottom of the trap and exhausted by the fan. BG-Sentinel devices were set in the lower level of the house, generally abutting a wall, but with a clear space directly above them to allow the air plume above the trap to flow freely; they were set between 1000 and 1400 h and were retrieved 24 h later.

The EVS trap (Australian Entomological Supplies, Bangalow, NSW, Australia) was baited with 1 kg of dry ice (CO₂), and deployment also was also 24 h.

Two unsuccessful methods were not included in the analysis. One method used a 12-cm-diameter × 12-cm-height cylindrical trap covered in an adhesive strip (Silvandersson Miljö AB, Knäred, Sweden) and contained 2 granular media CO₂ sachets (JW Hock). A 5-cm, 12-V electric fan pushed CO₂ out of the cylinder through 22 5-mm holes. The second method used a 23-cm-sweepnet operated for 10 min under a house, targeting *Ae. aegypti* harborage sites. The cylindrical trap was included originally but was substituted with the sweepnet after it failed to capture any mosquitoes. The two methods were used at 12 houses each.

Physiological state of *Ae. aegypti*

After identification in the laboratory, female *Ae. aegypti* were dissected under a microscope and classified as either nulliparous (coiled tracheolar skeins visible in the ovaries), recently bloodfed (blood in the midgut), gravid (ovarioles developed past stage II), or parous (absence of tracheolar skeins in ovaries) (Detinova 1962). Bloodfed and gravid females were not classified for parity because they were both considered potentially dengue-positive regardless of previous egg development. This classification system enabled comparison of the frequency of physiological states captured by the different methods.

Statistical analysis

Mean collection data were log ($x + 1$) transformed to normality before analysis of variance

Table 1. *Aedes aegypti* collection data gathered using three sampling methods in Cairns, Queensland, Australia.

| | CDC Backpack Aspirator | BG-Sentinel | EVS trap | Statistical result |
|--|------------------------|----------------|---------------|--------------------------------|
| Total <i>Ae. aegypti</i> | 70 | 112 | 21 | |
| <i>Ae. aegypti</i> ♀:♂ | 24:46 (0.52:1) | 46.66 (0.70:1) | 17:4 (4.25:1) | $\chi^2 = 14.69, P = 0.0006$ |
| Mean ¹ <i>Ae. aegypti</i> ♀/collection | 1.00 ± 0.35a | 1.92 ± 0.39b | 0.71 ± 0.27a | $F_{72,2} = 4.36; P = 0.017$ |
| Mean <i>Ae. aegypti</i> ♂/collection | 1.92 ± 0.58a | 2.75 ± 0.61a | 0.17 ± 0.08b | $F_{72,2} = 12.18; P < 0.0001$ |
| Mean <i>Ae. aegypti</i> /collection | 2.92 ± 0.89a | 4.67 ± 0.84a | 0.88 ± 0.32b | $F_{72,2} = 9.54; P < 0.0001$ |
| Total mosquitoes | 183 | 147 | 73 | |
| <i>Ae. aegypti</i> :other spp. (% <i>Ae. aegypti</i>) | 70:113 (38.3) | 112:35 (76.2) | 21:52 (28.8) | $\chi^2 = 63.58, P < 0.0001$ |
| No. <i>Ae. aegypti</i> positive collected | 17/24 | 19/24 | 8/24 | $\chi^2 = 4.68, P = 0.0963$ |

¹ Means ± SEM followed by different letters indicate statistically significant differences ($n = 24$ collections).

(ANOVA) with least significant difference (LSD) *post hoc* testing by using SPSS release 11.0.1 (SPSS Inc., Chicago, IL). Frequency data were analyzed using contingency tables and the chi-square (χ^2) statistic (Zar 1999), in which the null hypothesis of independence of sampling method was tested.

RESULTS

Mosquito collections

In total, 403 mosquitoes were collected in this study, 203 (50.4%) of which were *Ae. aegypti*. Other species were *Culex quinquefasciatus* Say (26.3%), *Cx. sitiens* Wied. (8.7%), *Ochlerotatus notoscriptus* (Skuse) (6.0%), *Oc. vigilax* (Skuse) (2.7%), *Verrallina funerea* (Theobald) (1.7%), *Cx. hilli* Edwards (1.5%), *Oc. kochi* (Donitz) (1.2%), *Anopheles farauti* Laveran s.l. (0.5%), *Cx. annulirostris* Skuse (0.5%), and *Uranotaenia* sp. (0.5%).

BG-Sentinel devices collected the most *Ae. aegypti*, followed by the CDC Backpack Aspirator and the EVS trap (Table 1). BG-Sentinel devices collected significantly more female *Ae. aegypti* than both alternatives ($P = 0.017$; Table 1). Male-only and combined male-female *Ae. aegypti* collections for the BG-Sentinel and CDC Backpack Aspirator

were significantly greater than collections in the EVS trap ($P < 0.0001$; Table 1). No positional effects on *Ae. aegypti* abundance were detected.

Both the CDC Backpack Aspirator and BG-Sentinel captured more males than females, significantly different to the male:female ratio of EVS traps that collected proportionally more females ($P = 0.0006$; Table 1). The BG-Sentinel was significantly more specific for *Ae. aegypti*, with 76.2% of its collection being this species, compared with approximately 30–40% for the two other methods ($P < 0.0001$; Table 1). The frequency of *Ae. aegypti*-positive collections was higher in the CDC Backpack Aspirator and BG-Sentinel than the EVS trap but not significantly higher ($P = 0.0963$; Table 1).

Of the unsuccessful methods not included in the analysis, the cylindrical trap with granular CO₂ media and adhesive did not capture any mosquitoes, whereas the sweepnet only captured 3 *Ae. aegypti* (2 males, 1 female), giving a mean *Ae. aegypti* collection of 0.23 ± 0.12 .

Concerning the next most common species, *Cx. quinquefasciatus*, the CDC Backpack Aspirator had significantly greater ($F_{72,2} = 10.66; P < 0.0001$) mean ± SE collections (3.2 ± 0.8) than the BG-Sentinel device (1.0 ± 0.5) and EVS traps (0.3 ± 0.1).

Physiological state of *Ae. aegypti* females

Dissections were performed on 74 of 87 (85.1%) female *Ae. aegypti* collected (remaining specimens were too dry or damaged). Analysis revealed that the frequency of physiological states collected varied significantly between sampling methods ($\chi^2 = 13.09, df = 6, P = 0.0416$). The CDC Backpack Aspirator collected more recently bloodfed specimens than the other 2 methods, which collected a greater proportion of nulliparous *Ae. aegypti* (Table 2).

Table 2. Percentage of female *Aedes aegypti* (actual number) in different physiological conditions, collected using three methods.

| Physiological condition | CDC | | |
|-------------------------|--------------------|-------------|----------|
| | Backpack Aspirator | BG-Sentinel | EVS trap |
| Nulliparous | 16.7 (4) | 42.1 (16) | 50 (6) |
| Parous | 25.0 (6) | 18.4 (7) | 25 (3) |
| Gravid | 16.7 (4) | 31.6 (12) | 8.3 (1) |
| Recently bloodfed | 41.7 (10) | 7.9 (3) | 16.7 (2) |
| Total dissected | 24 | 38 | 12 |

DISCUSSION

The BG-Sentinel and CDC Backpack Aspirator are effective *Ae. aegypti* sampling devices for use in northern Queensland. Based on trapping efficacy, both methods were superior to CO₂-baited EVS traps. The BG-Sentinel collected significantly more female *Ae. aegypti* than the CDC Backpack Aspirator, although both methods were equally efficacious when male-only or combined male-female collections were considered. The BG-Sentinel was also the most *Ae. aegypti*-specific sampling method.

The different methods do not sample similar components of *Ae. aegypti* populations. BG-Sentinel devices collected relatively more nullipars than the CDC Backpack Aspirator, which collected relatively more recently bloodfed females. This difference was probably due to the resting behavior of recently bloodfed specimens, with the aspirator targeting resting mosquitoes. Conversely, the BG-Sentinel collects flying mosquitoes, including young females in search of hosts and mates. Further research is required to determine which method best samples the entire age structure of natural *Ae. aegypti* populations. The development of techniques to determine the calendar age of individual *Ae. aegypti* (Gerade et al. 2004) will aid in this assessment.

The higher female:male ratio in EVS trap collections is most likely related to the use of CO₂ as an attractant, because this bait attracts female mosquitoes. BG-Sentinel collection devices use not only olfactory but also visual attractants, whereas the CDC Backpack Aspirator is nonattractant and does not discriminate against male collections.

The data presented here define the relative performance of three potential *Ae. aegypti* sampling methods in northern Queensland, Australia. Such data can aid researchers and public health workers in decisions concerning which *Ae. aegypti* sampling device will best suit their requirements. However, applying these results to *Ae. aegypti* in other regions will require careful consideration.

The choice of any method should consider factors such as local housing structure, cultural values, available funding, labor costs, and various scientific concerns. For example, the condition of *Ae. aegypti* specimens required may influence the choice of sampling method. If blood meal analysis is to be done, then a CDC Backpack Aspirator would be preferred because it collects proportionally more bloodfed females. Alternatively, if nullipars are needed, then a BG-Sentinel would be preferred. If the goal is to maximize the size of *Ae. aegypti* collections and funds are not limiting, then the BG-Sentinel would be the preferred sampling device. In addition, if house entry is problematic because of noncompliance by residents or for cultural reasons, then the BG-Sentinel may be more useful, because it can be set in a sheltered area immediately outside the house. Furthermore, prearranged permission to

set a BG-Sentinel outside of homes may negate the problems of house access during the working day when residents are not at home.

The BG-Sentinel offer a standardised collection method, whereas collections using the CDC Backpack Aspirator are influenced by variable operator performance. If neither device is available, the EVS CO₂-baited trap can be used to detect the presence of *Ae. Aegypti*, but it is not as sensitive as a CDC Backpack Aspirator or BG-Sentinel.

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