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# TAMING A TIGER IN THE CITY: COMPARISON OF MOTORIZED BACKPACK APPLICATIONS AND SOURCE REDUCTION AGAINST THE ASIAN TIGER MOSQUITO, *AEDES ALBOPICTUS*

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ABSTRACT. We evaluated 2 strategies to manage *Aedes albopictus*: 1) motorized backpack applications and 2) source reduction (coupled with hand-applied applications of larvicide). Backpack applications used a water-dispersible granular formulation (VectoBac® WDG) of *Bacillus thuringiensis* var. *israelensis* (*Bti*), whereas source reduction used granular formulations of the insect growth regulator methoprene (Altosid®) combined with a monomolecular film surfactant (Agnique®). Six subplots (total 8.02 ha) were selected for backpack applications, source reduction, and control groups. The experiments were blind with applications conducted randomly and independently. Efficacy was determined through placement of bioassay cups with larvae within experimental plots 1 day before treatment. Backpack applications resulted in 76% (±8.2% SE) and source reduction resulted in 92% (±4.1% SE) larval mortality. Backpack applications required 50 times less labor than source reduction (0.25 versus 0.005 ha/h). The cost of backpack applications, including labor, was $159.88/ha, compared with $659.65/ha for source reduction. Although overall efficacy was slightly lower, motorized backpack applications of *Bti* were more efficient and cost-effective than source reduction methods to control *Ae. albopictus* in urban settings at the community level.

**KEY WORDS** *Bacillus thuringiensis israelensis*, *Bti*, methoprene, hand applications, backpack applications

## INTRODUCTION

*Aedes albopictus* (Skuse) has spread to 36 states since it was 1st discovered in Texas, USA, during 1985 (Sprenger and Wuithiranyagool 1986, Enserink 2008). In New Jersey, *Ae. albopictus* has spread to 20 of 21 counties (Farajollahi and Crans 2012) since it was 1st detected in 1995 (Crans et al. 1996). This mosquito species is an aggressive human biter and a known vector of several arboviruses impacting veterinary and human health (Hawley 1998, Gratz 2004, Turell et al. 2005, Armstrong et al. 2013). The pestiferous behavior of this species, coupled with its potential public health concerns, make *Ae. albopictus* a top priority for control efforts (Farajollahi and Nelder 2009, Farajollahi et al. 2012, Rochlin et al. 2013).

*Aedes albopictus* suppression is difficult because the larval habitats of this peridomestic species are ubiquitous and cryptic within the urban setting (Bartlett-Healy et al. 2011, 2012; Unlu et al. 2013). Local mosquito control programs in urban settings have focused on source reduction in specific parcels within an area as a response to growing *Ae. albopictus*–related service requests (Farajollahi and Nelder 2009). In traditional source reduction, inspectors check the property for all potential mosquito habitats and remove them or apply larvicides as necessary. A major challenge is access restrictions (residents not home during work hours, vacant and locked properties, or access denial from private residents to government workers) where it can be difficult to remove or treat all potential habitats (Unlu and Farajollahi 2012).

Tremendous time and personnel resources are needed to suppress *Ae. albopictus* effectively, especially since effective control needs to be implemented on a large scale, at the community level. Source reduction, coupled with the use of insecticides, is feasible in a large area only if unlimited resources such as personnel and funding are available. However, the current trend of shrinking budgets limits available resources for mosquito control programs. Simultaneously, *Ae. albopictus* continues to expand its range to rural areas (Farajollahi and Nelder 2009), stressing current resources and control efforts. A necessity exists to explore alternative and more effective methods for area-wide control of *Ae. albopictus* in hopes of finding economically feasible and sustainable control strategies.

Backpack applications use a motorized backpack blower to mass spray insecticides through a misting nozzle. The equipment is relatively easy to carry and can deliver insecticidal mist to a

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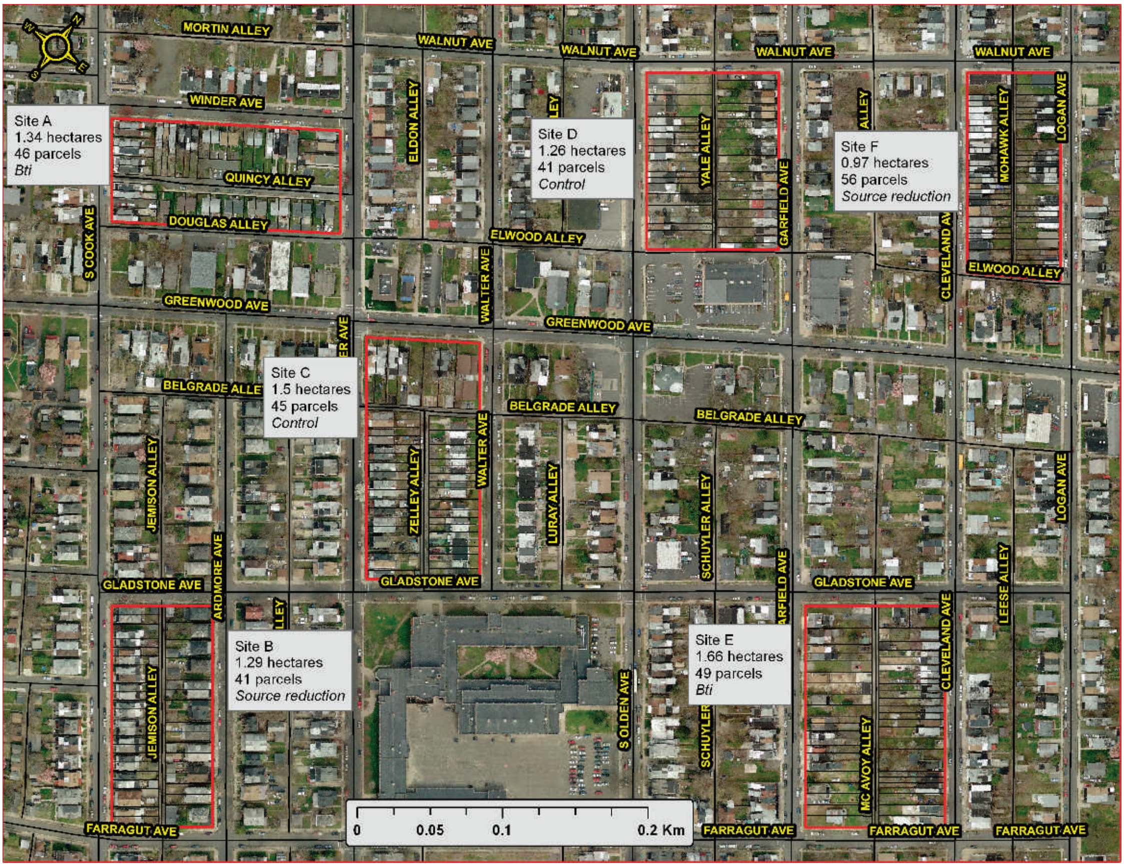

Fig. 1. Aerial photograph of field site subplots and parcels in Trenton, New Jersey. Subplots A and E were treated with *Bacillus thuringiensis* var *israelensis*; subplots B and F received source reduction; subplots C and D were the control groups.

large area in a short time. Backpack sprayers have been used internationally to control dengue vectors, both indoors and outdoors in suburban residential areas and temporary settlement sites in Malaysia (Lee et al. 2008), and for control of *Ae. albopictus* in natural habitats in Singapore (Lam et al. 2010).

We examined the efficacy and efficiency of backpack applications and compared this technique to a traditional source reduction strategy to suppress *Ae. albopictus* area-wide within urban settings.

## MATERIALS AND METHODS

### Study sites

We selected sites in the city of Trenton, Mercer County, NJ (40°13′N, 74°44′W) because of previous requests for service related to *Ae. albopictus* and the known abundance of this species during routine mosquito surveillance efforts (Fonseca et al. 2013). Our test site was a highly urbanized residential site with an area of 48.44 ha that included 1,251 residential parcels that comprise a housing structure and surrounding yard, and are often built as adjoining row homes or duplexes (Unlu et al. 2011). The site contains 26 residential blocks, each approximately 175 × 90 m. We selected 6 of these residential blocks (subplots A, B, C, D, E, F) to conduct our studies (Fig. 1). Subplots A (1.34 ha, 46 parcels) and E (1.66 ha, 49 parcels) were chosen for backpack applications; subplots B (1.29 ha, 41 parcels) and F (0.97 ha, 56 parcels) were chosen for source reduction; and subplots C (1.50 ha, 45 parcels) and D (1.26 ha, 41 parcels) were chosen as the control (no treatment) locations. Within each subplot, 20 parcels were selected at random for our experiments, and within each parcel 5 locations were chosen for bioassay cup placement (front of the home, side, porch, back yard, and alcove). In total, 278 individual parcels were sampled, covering 8.02 ha. Field crews asked permission from property owners to access each parcel prior to the experiment.

## Mosquitoes

Larvae used in all experiments were obtained from a colony of *Ae. albopictus* established from an automotive salvage yard located in Trenton, NJ (Nelder et al. 2010). Maintenance of the colony and rearing of larvae followed established protocols (Gerberg et al. 1994, Nelder et al. 2010). Briefly, egg papers were submerged in 2 liters of dechlorinated tap water (1 paper per enamel tray) containing 0.15 g of lactalbumin/brewer's yeast (1:1 ratio by mass), and eggs were allowed to hatch at 27°C under a 16:8 h L:D photoperiod. Egg papers with unhatched eggs were removed from trays after 24 h to ensure uniform hatch (i.e., experimental use of similar age larvae). The tray was cleaned daily as necessary. Larvae were fed on finely ground rat chow (0.5 g dissolved in 50 ml tap water) until they reached 2nd or 3rd instar. Only 2nd- or 3rd-stage larvae were used for subsequent assays.

## Bioassays

Three teams of 4 individuals each placed cups to be used for bioassays within experimental subplots the day before inspection and treatment efforts. The experiment was designed as blind; placement and inspection and treatment teams did not have contact during the experiment. The inspection and treatment teams were unaware of the designed study being undertaken as well. When possible, bioassay cups were not placed in open locations but were hidden along fences, under vegetation, beside or within garbage cans, toys, or discarded bottles, and in other habitats preferred by *Ae. albopictus*. Bioassays were performed by placing one 500 ml polystyrene wide-mouth container (Uline, Pleasant Prairie, WI) with 250 ml of dechlorinated tap water at each location within the experimental subplots (6 subplots × 20 parcels × 5 locations = 600 cups). Each cup contained 20 larvae, which were maintained in the laboratory as described above.

## Treatment applications

Backpack applications were conducted by a team consisting of 2 mosquito control inspectors. The inspectors were tasked with treating the entire 2 subplots (A and E) as they would during routine inspection and control efforts. The inspectors checked and treated any potential mosquito larval habitat within all the parcels (46 parcels in subplot A, 49 parcels in subplot E) contained in the 2 subplots. A Stihl® 420 backpack blower fitted with a liquid spray tank and a diffusor tip (Stihl USA, Virginia Beach, VA) calibrated to a flow rate of 0.12 liter/min was used. We used a Sympatec laser system (Sympatec, Clausthal, Germany) to conduct measurements of spray droplets and determined a $DV_{0.5}$ (volume median diameter) droplet size of 64.1 μm using an airspeed velocity of 23.5 m/s. The $DV_{0.5}$ is where 50% of the spray volume or mass is contained in droplets smaller than this value. The amount of time inspectors spent within each subplot was recorded.

Backpack applications used a water-dispersible granule formulation (VectoBac WDG®, Valent BioSciences, Libertyville, IL) of *Bacillus thuringiensis* var. *israelensis* (*Bti*) de Barjac containing 37.4% AI or 3,000 international toxic units per mg. The *Bti* powder was mixed at a dilution rate of 0.45 kg/3.79 liters of tap water and sprayed at an application rate of 0.49 kg/ha. Each parcel was divided in half, and each inspector sprayed all potential habitats in the front and back yards.

Source reduction was conducted by 2 teams of 4 individuals each. The personnel inspected each parcel (41 parcels in subplot B, 56 parcels in subplot F) and conducted source reduction by removing and discarding disposable containers (discarded tires, cups, boxes, food containers, etc.) that contained or could collect water. Container habitats that could not be removed (trash cans, bird baths, stored tires, tarpaulin covers, etc.) were hand-treated with pesticides. Altosid Pellets containing 4.25% AI of methoprene (Central Life Sciences, Schaumburg, IL) were placed in small containers such as trash cans and tires. Other nonremovable container habitats with an amorphous surface area, such as multiple pockets within tarpaulin covers, were treated using Altosid XR-Granules containing 1.5% AI of methoprene (Central Life Sciences). Altosid Pellets were applied at a rate of 2.72 kg/ha and Altosid XR-Granules were applied at 5.68 kg/ha (Nelder et al. 2010). When pupae were detected in larval habitats, a monomolecular film containing an alcohol-ethoxylated surfactant was used in all container types. Agnigue MMF Granules containing 32.0% AI (Cognis Corporation, Cincinnati, OH) were applied according to recommended label rates of 15.82 kg/ha. All containers in each parcel that were either holding water or had the potential to hold water after a rain event were either treated with pesticides or removed via source reduction as determined by inspectors. For each parcel, the amount of insecticide used was recorded in addition to the time inspectors spent within each parcel.

## Efficacy determination

Bioassay cups were retrieved 24 h after treatment and transported to the laboratory at a temperature of 25°C. Mortality rates of larvae in both control and backpack treatment cups were monitored for 72 h postretrieval. Efficacy of source reduction was recorded as 100% for a given location (front, side, porch, back yard, and

Table 1. Summary of insecticide usage and costs to control *Aedes albopictus* within treatment plots located in Mercer County, New Jersey, during 2009.

| Type of application | Insecticide | Amount (g) | Price per g ($) | Chemical cost ($) | Total chemical cost ($) | Labor cost ($) | Total cost ($) | Cost per ha ($) |
|---|---|---|---|---|---|---|---|---|
| Backpack | VectoBac WDG | 5,443 | 0.06 | 325.80 | 325.80 | 153.84 | 479.64 | 159.88 |
| Source reduction | Altosid Pellets | 74 | 0.05 | 3.93 | 7.12 | 1,483.70 | 1,490.82 | 659.65 |
| | Altosid XR-G | 91 | 0.02 | 1.56 | | | | |
| | Agnique MMF-G | 295 | 0.01 | 1.63 | | | | |

alcove) if the bioassay cup was removed or emptied by treatment teams. In cases where habitats were suspected but no larvae were detected, source reduction crews applied the Altosid and Agnique combination. If Altosid was detected in a bioassay cup, efficacy was also recorded at 100% because previous studies have shown that Altosid Pellets suppressed *Ae. albopictus* in a similar size container for over 100 days (Nelder et al. 2010). Larval mortalities in source reduction treatment plots were calculated based on the number of cups removed or treated, divided by the number of total cups placed.

An ANOVA and Tukey HSD statistical analysis of mortality within all parcels was performed using IBM SPSS Statistics 21 (IBM, Armonk, NY) to determine if there were significant differences among backpack applications, source reduction, and control groups and if there were significant differences within backpack applications and source reduction, i.e., if there were biases toward 1 location over another inside a parcel.

### Cost comparison

Economic comparisons were calculated on completion of the experiments. For backpack applications, the total time was calculated by the hours spent within subplot A and subplot E. The total amount of *Bti* ($0.06/g) used in subplot A and subplot E was summed and costs were calculated. For source reduction, the total time was calculated by the time spent on subplot B and subplot F. The total amount of Altosid Pellets ($0.05/g), Altosid XR-Granules ($0.02/g), and Agnique MMF Granules ($0.01/g) used in source reduction was calculated by adding the amount used in subplot B and subplot F, and overall costs were calculated. An application efficiency rate was calculated based on the amount of time that 1 mosquito control inspector spent to treat 1 ha. The labor cost was calculated based on time spent within plots, using $25.64/h (fringe benefits included) as the average cost per inspector.

## RESULTS

We used 5,443 g of *Bti* to treat subplots A and E with a backpack blower during our experiments for a pesticide cost of $325.80 (Table 1). The inspectors assigned with this task spent 80 min within subplot A and 100 min within subplot E, resulting in a labor cost of $153.84. Thus, the total costs for labor and pesticides used in backpack treatment plots were $479.64. We determined an application efficiency of 0.25 ha/h using backpack applications. Average larval mortality after 72 h for both *Bti* subplots was 76% (84% in subplot A and 68% for subplot E) (Fig. 2). Overall larval mortality in backpack application sites was significantly higher than that in the controls ($F = 452.664$; df $= 533$; $P < 0.05$); however, larval mortality of backpack applications was significantly less than that in the source reduction sites ($F = 73.305$; df $= 386$; $P < 0.05$). Larval mortality in each location inside a parcel (front, side, alcove, porch, and back) was calculated to be 92.42%, 69.71%, 62.28%, 72.87%, and 85.25%, respectively. Larval mortalities at front, side, alcove, porch, and back were not significantly different from each other except that the larval mortality at alcove was significantly less than that at front ($F = 3.749$; df $= 187$; $P < 0.05$).

In the source reduction treatment plots, we used 74 g of Altosid Pellets, 91 g of Altosid XR Granules, and 295 g of Agnique MMF Granules for a total pesticide cost of $7.12 (Table 1). The 8 inspectors assigned to this control method spent a total of 3,472 min conducting source reduction in subplots B and F, resulting in a labor cost of $1,483.70. In addition to pesticide applications, inspectors also removed 240 containers from subplot B and 88 containers from subplot F. The total costs for labor and pesticides used in the source reduction were $1,490.82. We determined an application efficiency of 0.005 ha/h using source reduction. Average larval mortality for source reduction plots was 92% (88% in subplot B and 96% for subplot F) (Fig. 2). The larval mortality in source reduction plots was significantly higher than that in the control ($F = 0.233$; df $= 199$; $P < 0.05$) and backpack application plots. Larval mortality for each location was 90%, 92.50%, 92.50%, 95%, and 90%, respectively, with no significant differences among them.

Larval mortality in control plots C and D was 0.175%. There were no significant differences among the locations.

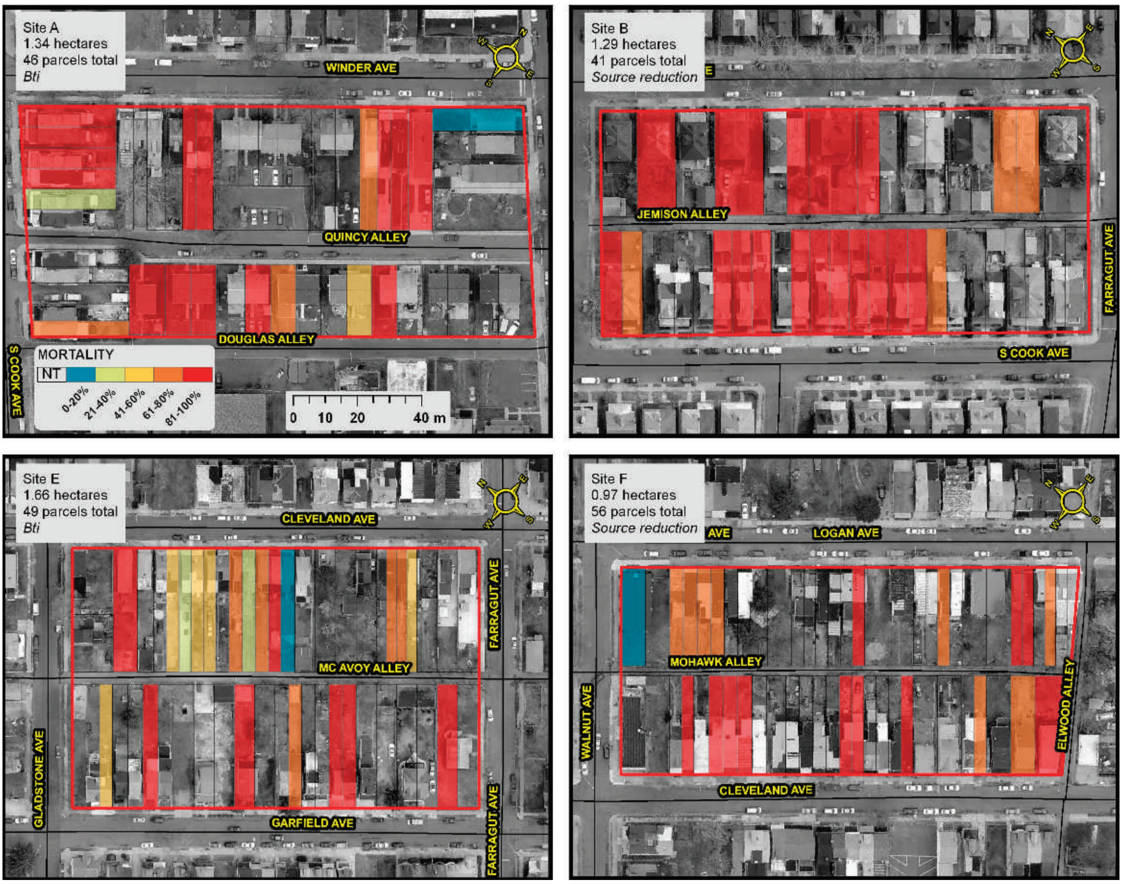

Fig. 2. Selected treatment subplots and parcels (in color) in Trenton, New Jersey, showing the percentage of *Aedes albopictus* larval mortality within treated parcels.

## DISCUSSION

Larval mortalities in backpack applications and source reduction were both higher than that in the control group, showing that both methods are effective at controlling immature *Ae. albopictus*. Both source reduction and use of insecticides play an important role in integrated mosquito management programs when targeting *Ae. albopictus*, but the true challenge lies in developing suppression techniques that are effective, efficient, economically sound, and do not exacerbate the problem of shrinking resources available to local government agencies tasked with mosquito control. Our results indicate that backpack applications of *Bti* can be used to suppress immature *Ae. albopictus*, especially in neighborhoods with a large number of containers that cannot be addressed by source reduction because of limited personnel. This method drastically reduces labor costs otherwise spent on identifying, treating, and removing larval habitats and can be an efficient method of quickly treating large areas.

The removal of mosquito oviposition habitats can be an effective method for mosquito control, but it requires community participation for sustainability (Winch et al. 1992, Gubler and Clark 1996, Bartlett-Healy et al. 2011). Furthermore, some potential habitats such as trash cans, tarpaulins, and flower pots are nonremovable because they are in use by homeowners. Unlu et al. (2013) found that immature *Ae. albopictus* are consistently found in plant saucers, buckets, and tires even after intensive source reduction practices. This makes the already-labor-intensive and therefore costly source reduction strategy more unaffordable and unfeasible. Consequently, this method may not be the best approach for area-wide control of a container-inhabiting mosquito species that continues to expand its range (Farajollahi and Price 2013). In addition, the cryptic and varied habitats found across urban landscapes make it difficult to detect larval sources, or to even access them in privately owned parcels. Professionals need better control techniques that can be applied quickly and effectively, with a reasonable cost to large areas that would successfully impact mosquito populations.

Larval mortality from backpack applications was 16% lower than that from source reduction in our experiments. For backpack applications, applicators sprayed a variety of habitats, including areas that were covered by thick vegetation while source reduction inspectors screened the potential habitats and checked for mosquito habitats. Although the bioassay cups placed in our experiments were relatively easy to find, pick up, and empty, they may have been difficult for a broad spray to penetrate at times. These factors are presumed to explain the differences in larval mortalities between the 2 management strategies.

Additionally, in source reduction, the larval mortalities among the 5 locations did not display any significant differences. This indicated source reduction could evenly control mosquito larvae in residential parcels. This was not a surprise because field crews were trained to check all possible areas and every container, regardless of presence of water, to eliminate mosquito larvae and the containers. In backpack applications, however, the alcove location had significantly lower mortality than the front location. Alcoves tended to be covered by vegetation and trash, which may have prevented the pesticide from penetrating hidden containers.

Previous studies have shown that *Bti* is well suited for control of *Ae. aegypti* (L.) in container habitats (Lacey 2009), providing 75 to 100% mortality against this species (Lima et al. 2005, Lee and Zairi 2006). Repeated backpack applications of *Bti* in natural habitats with dense vegetation also demonstrated good efficacy against *Ae. albopictus*, resulting in 66% reduction in ovitrap index and 80% reduction of larval density (Lam et al. 2010). We observed 76% mortality when sites were treated by backpack sprayers in a residential area with 1 treatment, which showed satisfactory efficacy and achieved a larval mortality rate similar to others.

Another area of concern with source reduction strategies for *Ae. albopictus* suppression is the labor requirement. We found that the total time spent in subplots treated by backpack applications was much less than the time spent on subplots treated by the source reduction strategy. The application efficacy of backpack applications was 0.25 ha/h, which is 50 times more than that of source reduction (0.005 ha/h). Source reduction was significantly more expensive than backpack applications when labor cost (99.52%) and chemical cost (0.48%) were combined. This economic saving would hold even if multiple applications of *Bti* using backpacks are needed. With shrinking budgets and personnel, combined with so much time spent on source reduction, it is unlikely inspectors will routinely be able to perform effective source reduction on a large scale.

Although *Bti* has shown its efficacy in a variety of habitats with little persistence in the environment (Lacey 2009), some studies indicate that a residual effect can occur even under field applications (Lima et al. 2005, Sumanadasa et al. 2011, Farajollahi et al. 2013). According to Sumanadasa et al. (2011), direct applications of *Bti* (VectoBac WG) in containers against *Ae. aegypti* provided residual efficacy that ranged between 1 and 3 months, depending on temperature and rainfall. Lee et al. (2008) reported that after 4 wk of misting with *Bti* (VectoBac WG) against *Ae. aegypti* and *Ae. albopictus* in a suburban residential area and a temporary settlement site in Malaysia, the ovitrap index decreased by at least 50%. This indicates that backpack applications of *Bti*, as in our study, especially with repeated applications, may have a residual effect that will help reduce oviposition habitats of *Ae. albopictus* in the future and provide a relatively longer control interval. In future studies, we will conduct multiple *Bti* applications and determine whether this approach provides a residual effect as well as a subsequent impact on adult mosquito populations.

In conclusion, backpack applications of *Bti* are an effective and efficient control method against *Ae. albopictus* over large areas. This method provides a distinct advantage over source reduction strategy in labor, cost, and time on a communal scale within urban areas.

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