OPERATIONAL NOTE

MR. MISTER: ROCKIN' THE AEDES OF THE SAN FRANCISCO BAY SALT MARSHES

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ABSTRACT. Mist sprayers (MS) are being rapidly adopted nationwide for applying larvicides to control peridomestic *Aedes aegypti*. Because MS can loft large quantities of larvicide over relatively long distances, we examined its efficacy in a tidal marsh habitat for controlling *Ae. dorsalis*. Liquid Vectobac 12AS larvicide, containing *Bacillus thuringiensis israelensis*, was applied at 1.16 liter/ha using a MS. Cards that change color when exposed to liquid were placed perpendicular to the path of the MS showed that the larvicide mist traveled up to 60 m from the MS and did not extend to 90 m. Use of the MS enabled a 4-fold increase in total hectares treated during 2020–21 relative to the prior 2 years without an increase in staff time. Notably, there was 83% reduction in the quantity of *Ae. dorsalis* larvae at 5 days posttreatment. Similarly, there was 63% reduction in adult female *Ae. dorsalis* that were collected in encephalitis virus surveillance traps from nearby communities relative to the prior 2 years. There were 2.3-fold fewer requests for service to address a mosquito problem from residents of communities that abut the tidal marshes, suggesting the applications had a positive impact on these communities. The MS offer an attractive alternative to hand treatments in tidal marshes where the use of unmanned aircraft or all-terrain vehicles is prohibited by national wildlife refuge managers.

KEY WORDS Aedes dorsalis, larval control, mist sprayer, salt marsh, service request

The San Francisco Bay is home to 7.8 million people, with more than 20% residing in Alameda County. The highest population densities occur in communities that abut the San Francisco Bay where marshes provide wildlife habitat, recreation for people, and mosquito production in the absence of control measures. The flight radius of several mosquito species from salt and bulrush marshes extends tens of miles, putting people living in the most populous regions of Alameda County at risk for bites and arbovirus infection. Aedes dorsalis (Meigen), the summer salt-marsh mosquito, reproduces exclusively in salt marshes and may contribute to the transmission of Jamestown Canyon virus in coastal California (Fulhorst et al. 1996). It is a particularly aggressive daytime and crepuscular biter that may fly upwards of 5 mi (8 km) in search of a blood meal (Verdonschot and Besse-Lototskaya 2014) before returning to the salt marsh to oviposit on Salicornia pacifica (Standl.), commonly known as pickleweed. Exceptionally high tides (i.e., king tide) trigger egg hatch, resulting in upwards of a 16-fold increase in the abundance of Ae. dorsalis larvae (Kramer et al. 1995). While much of the land that rings the San Francisco Bay is environmentally suitable for S. pacifica, much of the habitat has been degraded or lost due to the construction of gray infrastructure such as roads and rooftops. Approximately 10,000 acres (5,000 ha) of salt marsh remain around the San Francisco Bay. Water circulation channels were placed in salt marshes during the early 1900s to

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promote the outflow of tidal waters from the marsh and limit mosquito production. Environmental regulations limit the extension of water circulation channels, leaving the application of insecticides as a more practical alternative if salt-marsh mosquitoes are to be controlled. Larvicides are typically applied by mosquito control agencies shortly before or after an exceptionally high tide by foot, amphibious vehicle, or aircraft. Although the latter can cover hundreds of acres in a day, they are costly to purchase, maintain, or hire. For example, an aircraft mission to apply insecticide can cost in the upwards of \$20,000, while a truck-based application in a salt marsh is less than \$1,500 a day (approximately 10%) the cost). Truck-mounted mist sprayers (MS) that can loft a large quantity of larvicide several hundreds of meters have been adopted to control Ae. aegypti (L.) that reproduce in residential backyards. We sought to determine if a MS could be repurposed from our urban mosquito control program to control Ae. dorsalis in a salt marsh after an exceptionally high tide.

The study was conducted at a salt marsh in Fremont, CA (GPS coordinates: 37.497395, -122.010363), that is bounded by a hard-packed levee and inundated with seawater from the San Francisco Bay via Coyote Creek. The frequency and magnitude of tides were determined by measurements provided by the National Oceanic and Atmospheric Administration at the San Francisco Gate. A Super Duty Mist Sprayer (A1 Mist Sprayers, Ponca, NE) that was mounted to the bed of a Ford F150 truck was used throughout the study. The MS

was equipped with the Micronair AU5000 atomizer (Micron Sprayers Limited, Bromyard, United Kingdom). Calibrations were set with an operating bypass pressure of 345 kPa (50 psi), engine speed at full throttle (3,500 rpm), yielding a flow rate of 5.7 liters (1.5 gal) per min. Adjusting the throttle or bypass pressure directly affected the flow rate. Wind speed and direction was measured using a Kestrel 550 Pocket Weather Meter (Boothwyn, PA). Vectobac 12AS larvicide containing Bacillus thuringiensis israelensis de Barjac (Valent BioSciences, Libertyville, IL) that was mixed with red dye FD&C Red #40 (Novact Corporation, Miami, FL) was applied at 1,160 ml/ha (1 pint/acre) using a MS with the output nozzle directed 5-15 degrees below horizontal at a ground speed of 24 kph (15 mph) from a levy that was elevated approximately 1.5 m (5 ft) above the tidal marsh where surface wind speed was 17.2 kph (10.7 mph). The MS wind that propelled the Vectobac 12AS mist was 158 kph (98 mph) at the mister output atomizer.

Droplets of Vectobac 12AS mist were captured on 2 types of cards to determine if the droplet density or size that was captured on the 2 card types differed. Water-sensitive spray cards (TeeJet, Springfield, IL) that change color from yellow to blue when exposed to liquid or white index cards $(3 \times 5 \text{ in}, [7.6 \times 12.7 \text{ in})$ cm]) that showed the red-dyed droplets were affixed to wooden stakes. They were subsequently placed perpendicular to the path of the MS 0.5 m (1.5 ft) above the pickleweed canopy to capture the larvicide mist. The water-sensitive and white cards were placed next to each other at distances of 3, 7, 15, 30, 60, and 90 m (10-300 ft) from the levee edge at 4 sites along the drive path of the MS. Each group of cards was separated by approximately 0.6 km (0.4 mi) from one another along the length of the levee. The cards were collected within 15 min of the 12AS application, and each placed in a CD jewel case to prevent additional moisture from contacting the card or loss of the red-dyed Vectobac 12AS from the card. Cards were imaged using a Nikon D700 DSLR camera that was fitted with a Nikon AF-S Micro Nikkor 105mm 1:2.8G lens (Tokyo, Japan), and the droplet characteristics analyzed using ImageJ software (Schneider et al. 2012). The droplet size was calibrated in the ImageJ software by placing a microruler scale bar on each card prior to imaging (Drewes 2005).

A dipper (Catalog No. 1132BQ; BioQuip Products, Rancho Dominquez, CA) was used to collect mosquito larvae 1–3 days prior and 2–5 days after larvicide applications, as described previously (Hagstrum 1971). Encephalitis virus surveillance (EVS) traps (BioQuip Products) were baited with dry ice and a BG-Lure (Biogents, Regensburg, Germany), and placed overnight at sites within and adjacent to the study area to collect adult mosquitoes (N = 285EVS traps during 2018–19; N = 297 EVS traps during 2020–21). Mosquitoes were enumerated and identified to species using an Olympus SMZ800 dissection microscope (Olympus America, San Jose, CA). Public perception of mosquito abundance was assessed by measuring the number of calls for biting mosquitoes from people that lived within 4 km (2.5 mi) of the study area during the 2 years prior to and after the MS was used for mosquito control.

The Vectobac 12AS mist traveled up to 60 m from the mister, but it did not extend substantially beyond 30 m (Fig. 1A; 1 m is approximately 1 yd). Droplet density was greatest 7 m from the MS. Although a greater droplet density was observed using the watersensitive cards compared to the red-dyed 12AS droplets that were captured on the white cards, the difference was not significant (27.9 and 16.0 droplets/cm², respectively; unpaired *t*-test, P = 0.5591; Fig. 1A). Average droplet diameter measurements were largest on the red-dyed cards at 15 m from the mister (123.8 µm), and at 30 m for the water-sensitive cards (118.1 µm; Fig. 1B). However, across all distances from the MS, there was no significant difference in droplet diameter that was measured using the water-sensitive or white cards with red-dyed 12AS (unpaired *t*-test, P = 0.1352). Because there was not a notable difference between droplet density or area as measured using watersensitive cards or red-dyed 12AS on white cards, using either type of card should be suitable for quantifying droplet characteristics.

Many vector control laboratories report mean droplet diameter as mean droplet volume (DV_{0.5}). However, it may be of value to report the distribution of droplet area using a violin plot (Fig. 1C). The volume of Vectobac 12AS that was applied was calculated using the radius of droplets on yellow water-sensitive cards and the formula of a sphere (volume = $[4/3] \times [radius]^3 \times \pi$). When the calculated volume was converted to units indicated on the Vectobac 12AS label (pints/acre), we found that the application rate was optimal at 7–15 m from the mister (1.1 ± 0.4 pints/acre; Fig. 1D). Droplets likely evaporated as they traveled from the MS to the cards, resulting in the decrease in droplet diameter, area, and calculated volume (Fig. 1B, 1C, and 1D, respectively).

Use of the MS enabled a 4-fold increase in total acres treated during 2020-21 relative to the prior 2 years (1,528 ha [3,777 acres] and 373 ha [922 acres]), respectively; unpaired *t*-test, P < 0.0001) without a concomitant increase in staff time. Notably, there was an 84% reduction in the quantity of *Ae. dorsalis* larvae at 5 days posttreatment. Similarly, there was 63% reduction in adult female Ae. dorsalis that were collected in EVS traps from nearby communities relative to the prior 2 years (Fig. 1E). There was no significant difference in the number or height of high tide events during the 2 years of larvicide applications with MS and the prior 2 years that may have affected the number of Ae. dorsalis egg hatches (1.9 \pm 0.1 m [6.3 \pm 0.3 ft] for both periods, N = 266 and 267 high tide events; unpaired *t*-test, P = 0.4022). There were 2.3-fold fewer service requests (SRs) to address a mosquito problem from residents of



Fig. 1. Analysis and outcomes of Vectobac 12AS application from a mist sprayer (MS) used in a salt marsh that abuts the San Francisco Bay: (A) Droplet density, (B) diameter, (C) area, and (D) calculated volume of Vectobac 12AS at 3, 7, 15, 30, 60, and 90 m from the MS; (E) average number of adult female *Aedes dorsalis* collected in encephalitis virus surveillance (EVS) traps before and after MS was used to apply Vectobac 12AS in the study area; and (F) total number of service requests (SRs) from residents around the study area before and after the MS was used to apply larvicide. Error bars indicate standard deviation.

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communities that abut the tidal marshes during the study period relative to the 2 prior years (Fig. 1F), suggesting the applications had a positive impact on these communities. However, the analysis is limited in scope to the years of 2018–21. Had trapping efforts that were made during 2018–21 been conducted in prior years as well, a different outcome on adult *Ae. dorsalis* abundance and SRs may have been observed. Nonetheless, MS offer an attractive alternative to hand treatments in tidal marshes where the use of unmanned aircraft or all-terrain vehicles is prohibited by national wildlife refuge managers.

Our applications of the A1 Super Duty Mist Sprayer were conducted without adjustments to the equipment's original design. Awareness of wind speed and direction were key components of a successful application. Although we were pleased with these results, there are several mechanical modifications that may increase application efficacy. Adjusting the Micronair AU5000 blade pitch can alter the droplet size and density. Removing the Micronair AU5000 and reverting to customary flat or conical nozzles may provide a better spray pattern for applications from a levee into salt-marsh habitats.