

Aerial Biolarvicide Applications to Protect Hawaiian Forest Birds from Invasive Mosquitoes

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Introduction

Hawaiian honeycreepers (Fringillidae) are facing extinction due to invasive mosquito-borne diseases exacerbated by climate change¹. Urgent mosquito control targeting the vector, the Southern House Mosquito (*Culex quinquefasciatusis*) is critical.

We are trialing mosquito control methods, including application of biolarvicides. Here we summarize the 2023 trial by Zhao et al. (2024) on the aerial application of *Bacillus thuringiensis* israelensis (Bti) and Bacillus sphaericus (Bs), in forest bird habitats on Kaua'i and Maui, where four critically endangered honeycreeper species are struggling to survive. We also provide an update on strategies implemented since the trial.

Bti/Bs are naturally occurring bacteria that produce toxins lethal to mosquito larvae and are commonly used for mosquito control for human health². Aerial broadcast effectiveness and costs in Hawai'i were largely unknown prior to this. Our goals:

- Test efficacy by measuring impact on mosquito populations (trapping) and the reach of product droplets to the forest floor (mortality bioassay cups).
- Implement aerial broadcasting within critical habitat.

Methods for Application

- Certified, trained pilot and equipped helicopter. Landing area for mixing products and refueling. Helicopter flight tracking, pre-loaded flight maps, and boom sprayer with calibrated nozzles.
- Flights occur at 80–112 kph and ~30 m altitude, covering a 45 m spray width. Flow rate ranges from 56–105 L/min. Spray volume is 2 L/ha: 1.75 L/ha VectoBac 12AS (Bti) and 560 g/ha VectoLex WDG (Bs)².
- Equipment includes a large water reservoir (>550L), large and small containers, water pump, hoses, drill with mixing attachment, and protective equipment.
- Favorable weather and temperature conditions are essential for safe flights and effective droplet distribution. Applications must occur during dawn or dusk inversions to keep droplets near the ground. Pesticide application must follow label instructions.
- Applications begin every 2 weeks, shifting to every 3 weeks. August-January, targeting peak mosquito breeding.

Key Takeways

The trial developed an aerial biolarvicide application protocol for forest bird habitats in Hawai'i. It showed that biolarvicides reached mosquito habitat, killing larvae and reducing local mosquito populations. We estimated that each session cost US\$20k (145 ha using 4 refills of 340 l of product/water).

Limitations include weather sensitivity, high landscape-level costs, time-intensive monitoring with variable results that complicate interpretation, and the need for specialized equipment and technical flying skills due to challenging terrain.

Advantages over other mosquito control strategies include lower cost and no need for overflooding ratios, making it suitable for areas with high mosquito densities.

We suggest that mosquito suppression using biolarvicides can be an effective tool within an Integrated Pest Management (IPM) framework to reduce avian disease risk and conserve Hawai'i's endemic bird species.

6 honeycreeper species

2023 Trial

Sites: Remote (no vehicle access), rugged steep terrain, high canopy wet (>4,000mm/year) forest with deep gulches.

Kaua'i trial in Alaka'i Wilderness Preserve: Maui trial in Hanawi Natural Area Reserve: 5 applications, October-December 2023 4 applications, August 2023 - January 2024 145 ha, Elevation: 1400-1700m 109 ha, Elevation: 1220-1570m

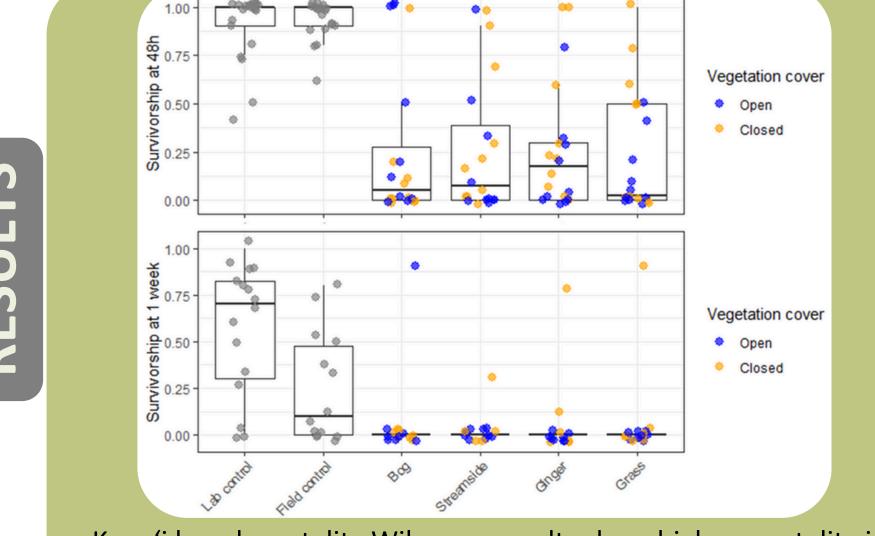
To assess how effectively larvicide droplets reached and performed, we used 6oz plastic cups as proxies for larval breeding habitats, placed within and outside treatment areas

Bioassay cups

Outside treatment: 10 cups placed in unobstructed habitat

Within treatment: 10 cups-5 obstructed, 5 unobstructed- each in 4 habitat types

Post application, cups were brought to the lab where 10 larvae and 100 mL distilled water were added. Live larvae were counted at 24 hr, 48 hr, and 1 week intervals to assess larval mortality³.



Kaua'i larval mortality Wilcoxon results show higher mortality in treatments than control (48h, p = 8.53e-14 and 1 Week, p = 3.86e-1409). Maui results: 48h & 1 week, p< 2.2e-16.

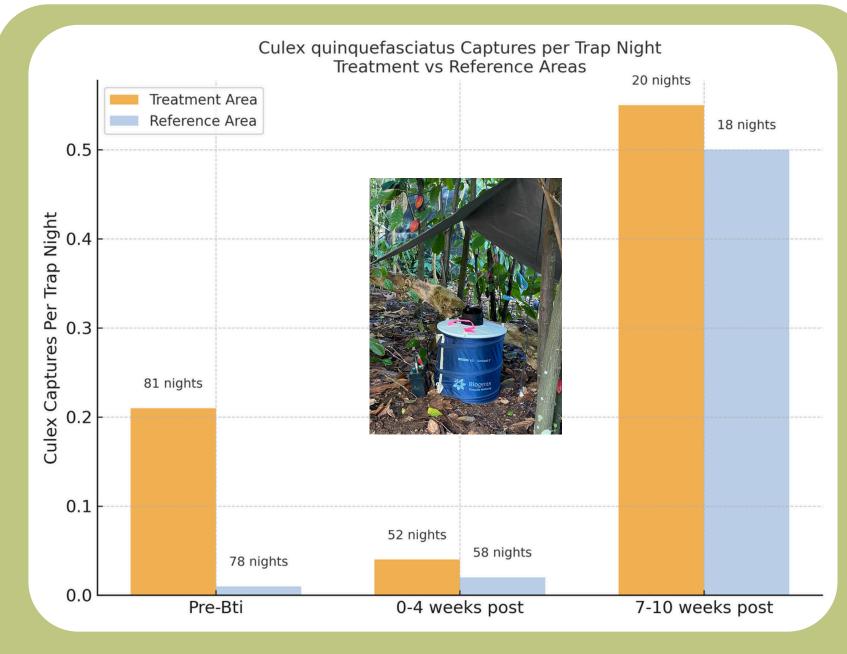
Results indicate effective product reach, even in dense vegetation and varied habitat. Variation in survivorship likely reflects weather conditions and differences in larval robustness.

8 honeycreeper species

To measure presence/absence of adult mosquitoes and evaluate efficacy, we deployed Biogents (BG) BG-Sentinel 2 traps designed to capture females seeking a blood meal.

Trapping (Kaua'i only)

5 traps/area (reference and treatment), 150-200 m apart in varied microhabitats, baited with BG Lure and CO₂. Trapping occurred for 2 nights/week before and during treatments, followed by biweekly trapping, when weather permitted. Post application: 4 nights.



41 mosquitoes in 307 trap nights over 6 months.

Results indicate reduced abundance in treatment vs. reference. Rates returned to pre-treatment levels once applications ceased, highlighting need for continuous application. Caveats- limitation of access, site terrain differences likely contributed to catch rates, relatively small mosquito population.

Current Strategy

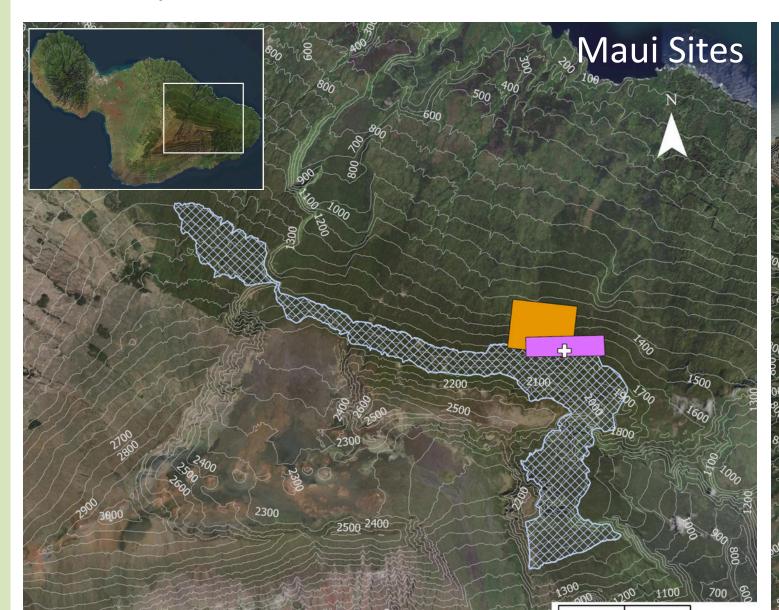
Due to the study findings, we expanded applications of biolarvicides. Modifications include using only Bti, which is more effective for initial knockdown than Bs, and Bs's extended longevity was deemed unnecessary due to frequent rain. More frequent applications are required, but the cost is less than a combined Bti/Bs application schedule. We also apply more concentrated droplets to use less water and reduce application time while delivering the same dosage rate. We are targeting areas that protect core populations of critically endangered honeycreepers.

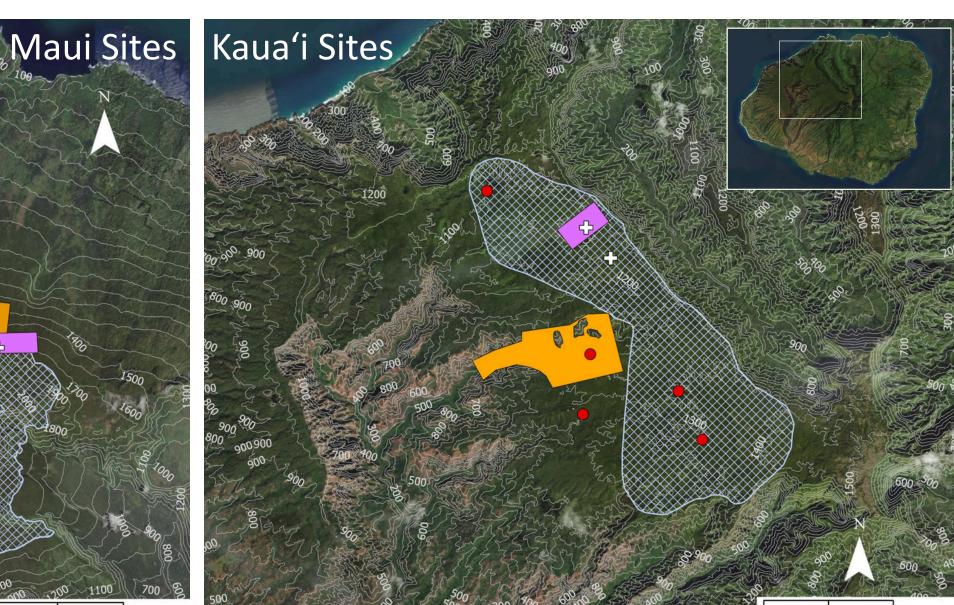
Maui updated application:

• August 2024-February 2025, applications occurred 1-2 times per week every 2 weeks on ~300 ha. Each session is 5-6 hours. No monitoring due to helicopter and access limitations.

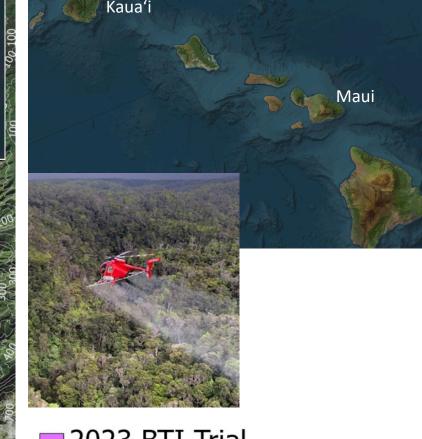
Kaua'i updated application:

• Since June 2024, >16 applications, 445 ha. Monitoring: Using bioassay cups and BG traps with yeast (to produce CO₂) in Bti treatment areas and a reference site. Results have been positive, larvae are dying in the cups.





From left to right: Current and past Maui Bti sites with inset map of location on Maui, Current and past Kaua'i Bti sites with inset map of location on Kaua'i, Maui and Kaua'i within the main Hawaiian Islands with legend and example of how Bti is sprayed from helicopter.



- 2023 BTI Trial
- 2024/2025 BTI Application
- 2024/2025 Monitoring Sites Endangered Bird Range Countours, every ~100m

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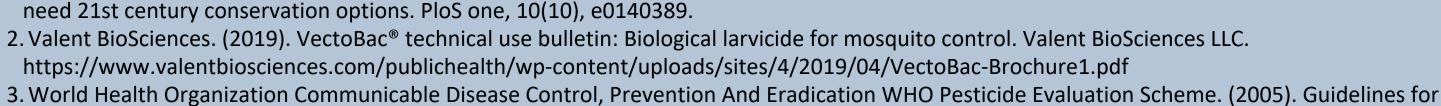


References

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1. Fortini, LB, Vorsino, AE, Amidon, FA, Paxton, EH, & Jacobi, JD. (2015). Large-scale range collapse of Hawaiian forest birds under climate change and the

