

Attractive toxic sugar bait stations for control of *Culex quinquefasciatus*

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INTRODUCTION

Sugar meals are essential to adult mosquito nutrition and only recently have been incorporated into systems that blend sugar bait with insecticides for mosquito abatement. Attractive toxic sugar baits (ATSBs) with multiple insecticides have been suggested to reduce resistance and enhance control agent efficacy against multiple mosquito life stages. We report here on laboratory and field assessments of ATSB stations with the insect growth regulator pyriproxyfen (PPF) and the entomopathogenic fungus *Beauveria bassiana* (Bb) against *Culex quinquefasciatus* mosquitoes.

METHODS

ATSBs with different PPF and Bb contact zones were tested on host-seeking or gravid female mosquitoes feeding on attractive bait. Autodissemination of PPF/Bb by contaminated adults to glass bowls holding early fourth instar larvae was assessed. Exposure periods were 2 d in mesh cages (laboratory) or 5 d in mesh enclosures (field). Post-exposure, larvae were reared for up to one week to assess emergence rates and surviving adults transferred to rearing chambers were monitored for mortality for up to 21 days. *B. bassiana* infection was identified from adult mosquito cadavers incubated in 24-well plates in a high humidity box for up to one week. In gravid adult assays, egg rafts were enumerated and the total number and hatching rate of eggs were determined. The persistence of mosquitocidal effects was examined for different ATSB designs aged up to 2 weeks under laboratory or field conditions. In the field, *Cx. quinquefasciatus* from laboratory colonies were released within replicated pyramidal mesh PVC enclosures on top of fiberglass tubs (total height = 2.5 m), and dead individuals were collected daily over a 5-day period and live individuals were removed from enclosures on day 5. Each enclosure contained a single hanging ATSB with either a combination of PPF/Bb or water control and glass water bowls with larvae. Two release/recapture trials, one with fresh and one with aged (1 or 2 weeks) ATSB stations, were performed in spring and autumn of 2017. The spring trial included host-seeking

adult females ($n = 75$) and fourth instar larvae ($n = 75$, three glass bowls) and the autumn trial involved gravid adult females ($n = 40$) with fourth instar larvae ($n = 40$, two glass bowls).

RESULTS

Laboratory results linked PPF (with or without Bb) to significant larval and adult mortality compared to controls. Average emergence was reduced by 90% (fresh) to 50% (1 week aged) and average adult mortality ranged from 30-75%. ATSB designs that stored more PPF per station and resisted desiccation for longer periods were more effective overall against adults and larvae. Compared to PPF alone, the Bb/PPF combination produced slightly enhanced adult mortality when the ATSB was aged for a week. Bb used alone in an ATSB station was a potent delayed-onset adulticide against host-seeking females, producing greater than 60% average mortality and infection rates over a 2 week post-exposure period; however, Bb lethality against gravid females averaged less than 20%, with high rates of sublethal infections evident post-mortem. Sugar deprivation may be a key factor in determining PPF/Bb ATSB efficacy. Host-seeking adults sugar-starved for 24 hours displayed higher mortality and infection rates than non-starved cohorts. The two field trials resulted in similar trends for mosquito mortality: PPF/Bb enclosures greatly reduced adult ($86 \pm 11\%$) and larval ($59 \pm 12\%$) abundance compared to controls (adults: $21 \pm 17\%$ and larvae: $16 \pm 13\%$). Field-acquired Bb infection was detected in an average of 10% of recaptured adults. Egg raft production by field-exposed gravid adults was reduced, on average, by nearly half in PPF/Bb enclosures (15 ± 9 rafts) compared to control enclosures (28 ± 3 rafts). Overall, an ATSB with pyriproxyfen and *B. bassiana* provided sustained control of adult and larval *Culex quinquefasciatus* and future field deployments targeting wild populations are the next step to further determine the usefulness of this system for mosquito abatement operations.