Inexpensive Trap for Monitoring the Green June Beetle

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ABSTRACT Green June beetle, *Cotinis nitida* (L.), is an important pest of grapes, peaches, blackberries, blueberries, apples, and pears. Currently, there is no inexpensive, commercially available lure or trap that could serve monitoring green June beetle adults. The objective of this study was to develop and optimize an inexpensive bottle trap baited with isopropanol to attract and capture green June beetle adults. Bottle traps baited with 8 mm diameter cotton wicked dispensers emitted from 9 to 43 ml isopropanol in 48 h and maintained that alcohol at a fairly constant concentration compared with the prototypical bottle trap with large surface evaporation of isopropanol poured into the bottom of the trap. Over 5 d, the isopropanol in the wicked dispensers remained at the same stable concentration of 45–44.5%, whereas isopropanol concentration in the bottom of prototypical traps dropped from 45% to ≈11% after 24 h and to 0.2% by 48 h. Bottle traps with isopropanol dispensers and cotton wicks of 4, 6, or 8 mm in diameter caught significantly more green June beetles than did prototypical bottle traps with no dispensers. Isopropanol concentrations of 45.5, 66, and 91% attracted more green June beetle adults than the lower concentrations. Significantly more green June beetle adults were attracted to traps with dispensers set at 1.3 m height than those at lower heights, and traps topped with a blue, orange, or white band captured more green June beetle adults than those with bands of other colors. The optimized bottle trap is made from recycled transparent polyethylene terephthalate beverage bottle (710-ml; 24 oz.) with a blue, orange, or white band, baited with an 8 mm cotton wick dispenser of 45.5% isopropanol and hung at a height of 1.3 m. Cost and uses for this trap are discussed.

KEY WORDS Coleoptera, Scarabaeidae, isopropanol, kairomone

Green June beetle, *Cotinis nitida* (L.), is an important pest of grapes, peaches, blackberries, blueberries, apples, and pears. The grubs of this species live and pupate in the soil. Typically, adults emerge, mate, lay eggs, and feed in mass on ripening and/or overripe fruit from mid-June through August (Iftner 1978). However, because of drought from early June to mid-August 2011 in north-western Arkansas (<1 in. rain each month), emergence of some local populations of green June beetle adults was delayed until early to mid-August and those adults continued to be caught in traps into September (D.T.J.).

Newly emerged, virgin green June beetle females release sex pheromone near their emergence holes in turf or later as they rest in the canopy of trees adjacent to the area of emergence. Green June beetle males search for mates by flying low over such areas or around tree canopies (Brandhorst–Hubbard et al. 2001). Mated females lay eggs in areas with decomposed manure or vegetative matter (Domek and Johnson 1988, Flanders and Cobb 2000). A week or two after emergence, green June beetle adults start searching for and feeding on ripe and overripe fruit. These fruit become inoculated with yeast and fungi that causes fruit to ferment and release volatiles that elicit green June beetle aggregation behavior (Domek and Johnson 1988, 1990; Vishniac and Johnson 1990; Johnson et al. 2009). Sometimes, all the pulp of a fruit or cluster of grapes is consumed and adjacent uneaten fruit get covered with beetle excrement rendering fruit unmarketable. In fall 2007, researchers from University of Arkansas surveyed county extension personnel and growers about the pest status of green June beetle in Alabama, Arkansas, Georgia, Missouri, Mississippi, New Jersey, North Carolina, Oklahoma, South Carolina, Tennessee, and Texas. The green June beetle was reported to reduce yields annually in a total of ≈13,100 ha of fruit and turf across these states with yield losses of at least $3.6 million (Johnson et al. 2009).

Management tactics against the green June beetle have changed over the last century with changes in nutrients (manure or synthetic fertilizer) applied and landscape use. Until the early 20th century, manure or decayed vegetable matter was a common source of nitrogen and other nutrients to maintain soil quality

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and fertility of many row crops and turf. As a result, there were many reports of green June beetle grubs causing indirect damage to turf in golf courses and many row crops (Chittenden and Fink 1922). Grub damage was reduced by making evening insecticide applications to kill the nocturnal grubs feeding near the surface (Davis and Luginbill 1921, Chittenden and Fink 1922, Flanders and Cobb 2000). As synthetic fertilizer replaced manure, there were fewer reports of grub damage to row crops and turf. However, Miner (1951) reported an increase in local green June beetle populations and ripe fruit damage by green June beetle adults as more acres of fruit were planted adjacent to ever increasing acreage of livestock pastures and land treated with chicken manure. Currently, green June beetle causes problems predominantly as a fruit pest.

The current management tactic recommended to large scale fruit producers is to minimize mass attack of ripe fruit by green June beetle adults with insecticide sprays, but only a few recommended insecticides have an acceptable preharvest interval <3 d (Pontasch and Knutson 2010). Fruit growers need an alternative to spraying insecticides such as attract and kill strategy. The other recommended tactic calls for timely harvesting of ripe fruit and removal of rotting fruit to lessen attraction to and damage by green June beetle adults (Pontasch and Knutson 2010). In addition, small scale producers have used floating row covers or exclusion nets to prevent green June beetle feeding damage (Strang et al. 1992, Lesoing, 2011). Proper timing of those cultural control measures against green June beetle requires an inexpensive and effective tool for monitoring the presence and population dynamics of this insect. In either case, development of a bait and trap that is more attractive to green June beetle adults than a planting of ripe fruit is required.

Several lures have been evaluated for attractiveness to green June beetle adults. Anecdotic evidence from the literature indicates that the first attempts of creating green June beetle lure were made by Muma (1944), who used 100% caproic acid. Wylie (1969) captured thousands of green June beetle adults with a very complex and difficult to standardize mixture of yeast fermenting molasses. A mixture of phenylacetaldehyde, 2-phenylethanol, methyl-2-methoxybenzoate, limonene, and methyl salicylate (formulated by Trécé Inc., Adair, OK), similar to a patented Mix-M blend (Lopez et al. 2002) that attracts various scarab beetles and green June beetle, was both reported to be very attractive to green June beetle adults (Johnson et al. 2009). This scarab lure could be obtained from several companies for \$7/lure/wk or \$28/mo to keep a trap baited to monitor green June beetle seasonal flight. This lure has been used in trap designs that cost from \$7 to \$25. Available green June beetle monitoring traps are still expensive and that may limit their use.

Fifty percent isopropanol in bucket and vane traps was reported to be attractive to green June beetle adults and the flower scarab, *Euphoria sepulchlaris* (F.) in Florida and could be used as an inexpensive lure for green June beetle adults (Landolt 1990). More recently, Pszczolkowski et al. (2008) used 45.5% isopropanol (isopropyl alcohol available in of many stores) for collecting green June beetle adults in a study on green June beetle sexual dimorphism. Traps used in the study by Pszczolkowski et al. (2008) were made from transparent polyethylene terephthalate 710-ml (24 oz.) beverage bottles and were a modification of the traps originally designed by Dr. James Baker of North Carolina State University and previously proposed for collection of Asian ambrosia beetles, *Xylosandrus crassiusculus* (Motschulsky) by Bambara et al. (2002) and Oliver et al. (2004).

There is a body of evidence that color of the trap may affect its performance. For instance, Fleming et al. (1940) found that traps painted yellow attracted more Japanese beetles, *Popillia japonica* (Newman), than those painted white, blue, pink, red, orange, or green. Ladd and Klein (1986) reported, for the same insect, that white traps to perform the best, followed by blue, yellow, green, red, and black. Williams et al. (1990) showed that in another scarab, *Macrodactylus subspinosus* (F.), white traps were more efficient than yellow traps. In some Curculionidae black and brown traps perform better than yellow and white traps (Mizell and Tedders 1999). However, the authors are unaware of any study on effects of color on green June beetle trap efficacy.

This article describes an attempt to evaluate this inexpensive baited bottle trap prototype to attract and capture green June beetles. We performed field experiments to optimize trap design, isopropanol concentrations (lure), wick diameter of the dispenser, trap location in relation to the ground, and band color on the trap.

Materials and Methods

Study Sites. The studies were conducted in several sites: a commercial orchard of Red Haven peach (*Prunus persica* (L.) Batsch) (1.2 ha), a vineyard of Norton vines (*Vitis aestivalis* Michaux) (Vitaceae), (0.5 ha), or a vineyard of Catawba vines (*Vitis labrusca* L. × *Vitis vinifera* L.) all located on the Experimental Farm of Missouri State University in Mountain Grove, MO, and an apple (*Malus domestica* Borkh.) (2.4 ha) and peach (1.6 ha) orchard of several cultivars near Berryville, AR. The sprays that particular plots received are specified in descriptions of particular experiments.

Lure Prototyping. The lure consisted of various concentrations of first aid antiseptic and unscented 91% isopropyl alcohol (rubbing alcohol, Cumberland Swan, Smyrna, TN) purchased in pharmacies of local stores. Mixing equal amounts of isopropyl alcohol and distilled water resulted in a 45.5% solution of isopropanol referred to as "standard isopropanol lure." Additionally, in some experiments other concentrations of isopropanol in water were used.

We also compared attractiveness of rubbing alcohol (91% isopropyl alcohol by Cumberland Swan) with that of lure produced by mixing 910 ml of ASC grade isopropanol (produced by ChemProducts and distributed by Nurnberg Scientific, Portland OR) with 90 ml of double distilled water. To that end, seven traps

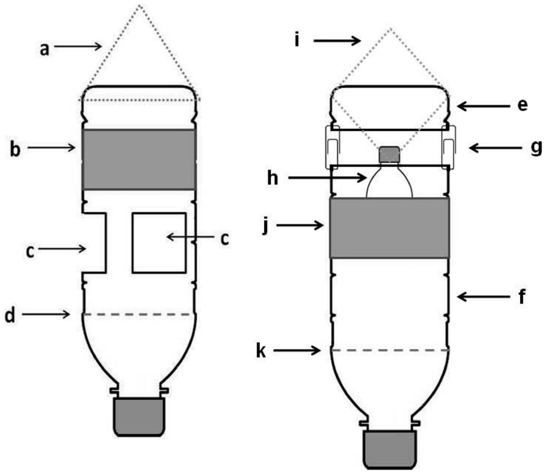

Fig. 1. The traps used in our study: (Left) standard bottle trap; (a) harness, (b) yellow band, (c) three square openings (the third opening is placed on the opposite side of the bottle), (d) maximum level of the lure; (Right) improved bottle trap; the bottom of the bottle (e) is cut off from the upper portion (f) and these two parts are reattached with paper clips (g). The isopropanol dispenser (h) is hung by a rope or wire harness (i), (j) color band, (k) maximum level of the killing agent (aqueous 0.02% Triton X).

baited with 91% rubbing alcohol and seven traps baited with 91% ASC grade isopropanol were randomly placed in unmanaged set aside plot in the Experimental Farm of Missouri State University in Mountain Grove, MO, in a distance larger than 500 m from the remaining experimental plots. The insects were collected after 48 h and identified according to Goodrich (1966). Green June beetle numbers per trap were calculated for traps baited with rubbing alcohol and for traps baited with ASC grade isopropanol and the averages compared with Student's *t*-test. Such experiment was performed with every new batch of rubbing alcohol purchased. In no case any statistically significant differences in attractiveness were observed between 91% rubbing alcohol and 91% ASC grade isopropanol. Thus, the attractive effect of rubbing alcohol is most likely because of isopropanol.

Trap Designs. The prototype called the "standard bottle trap" was made from recycled transparent beverage polyethylene terephthalate bottles (710-ml; 24 oz.) (Fig. 1a–d). Three square openings 4 × 4 cm for beetle entry were cut out from the wall of the bottle. The visual stimulus was a yellow polyethylene, opaque band (7-cm wide) that was stapled to the top of the trap. The odor stimulus was 125 ml of standard isopropanol lure poured into the bottle trap and also served as a killing agent.

The standard bottle trap was redesigned to accommodate a lure dispenser and is referred to as the "improved bottle trap" (Fig. 1e–k). The bottom 7 cm of the standard bottle trap was cut off and reattached to the top using three medium size paper clips. A 125 ml plastic bottle dispenser equipped with an 8 mm diameter cotton wick was filled with 100 ml of standard isopropanol lure. Alcohol was emitted through the space between the bottom and top parts of the bottle trap where attracted green June beetles entered the trap. This design had a 4-cm wide, yellow polyethylene, opaque band stapled to the bottle. The dominant wavelength, percentage reflected spectrum and CIE $L^*a^*b^*$ values for the band was determined by a Jaz spectrometer (Ocean Optics, Dunedin, FL), where $L^* = 0$ (black) to 100 (white); $a^* =$ negative (green) to positive (red); and $b^* =$ negative (blue) to positive (yellow) (Table 1). There was ≈200 ml of 0.02% Triton X-100 in water poured into the bottle as a killing agent. Before each use in the field, the traps were cleaned using scentless soap water, rinsed with distilled water, air dried, checked for damage, and mended as needed. In the following field experiments, all insects captured in each trap were emptied into a plastic bag, preserved with 91% alcohol, labeled appropriately, and placed in a refrigerator for later species identification and recording counts.

Table 1. Jaz Spectrometer (Ocean Optics, Dunedin, FL) measurements of the dominant color wavelength, percentage reflectance, and CIE $L^*a^*b^*$ values for each plastic color band used in traps

Color band	Dominant wavelength (nm)	% reflectance	L^*	a^*	b^*
Clear	—	—	—	—	—
White	Reflect all	88.0	95.7	−0.7	2.2
Red	625.4	50.0	39.9	58.4	26.9
Orange	587.0	53.0	61.0	26.6	55.0
Yellow	577.1	79.0	85.2	2.9	82.0
Green	545.3	35.0	56.3	−53.0	40.6
Blue	475.6	30.0	31.8	5.8	−45.9
Violet	425.0	37.0	34.2	53.2	−7.4
Black	Absorb all	0.5	3.9	9.2	19.8

Several experiments were conducted with the improved bottle trap to evaluate and optimize three factors for attractiveness to green June beetles including: the concentration of isopropanol emitted by the trap dispenser; the alcohol emission rate altered by changing the diameter of the cotton wick; trap placement; and the visual stimulus varied by stapling a different colored band to the trap.

Isopropanol Attractiveness. Seven standard bottle traps were filled with 125 ml of 0.02% Triton X in water (control) and seven were baited with 125 ml of standard isopropanol lure (experimental). All these traps were randomly located in the Norton vineyard at the height of 1–1.2 m. This plot was not sprayed with insecticides or fungicides but received herbicide sprays: one spray with Poast (sethoxydim) at 2.96 liters/ha in May and with a mixture of Surflan (oryzalin) and Gramoxone Inteon (paraquat) at rates 9.46 liters and 2.37 liters/ha, respectively, once in June and once in July. No fertilizers were applied in the Norton vineyard (Reut et al. 2010). The insects were collected after 48 h, identified according to Goodrich (1966), sexed according to Pszczolkowski et al. (2008) and green June beetle numbers recorded. This experiment was repeated weekly from 7 May to 9 August 2009.

Emission Rate. Four lure dispensers (125 ml), each equipped with a cotton wick of different diameters (2, 4, 6, and 8 mm), each had 8 mm length of wick protruding from the dispenser cap after being saturated with 45.5% isopropanol. Each dispenser was placed in the improved bottle trap. Additionally, one standard bottle trap was filled with 125 ml of 45.5% isopropanol. These five treatments were hung in a fume hood at temperature of 22 ± 2°C. The exhaust of the hood was adjusted so the air flow in the hood equaled 1 m/s. Every 24 h for five consecutive days, or until all lure had evaporated, measurements were made of the volume and percentage of isopropanol left in the trap. To measure the volume of evaporated lure, each dispenser with lure inside was weighed at the beginning of the experiment. Every 24 h each dispenser was reweighed and the actual weight of the dispenser subtracted from respective original weight. Next, the volumes of evaporated lure were calculated for each day and dispenser using isopropanol wt:vol standard curve. The percentage of isopropanol was measured by using a hydrometer that was calibrated for isopropanol using a standard curve of concentrations. This experimental procedure was repeated seven times.

These same five treatments were randomly assigned in the commercial peach orchard at Mountain Grove, MO. No insecticide or fungicide sprays were applied to this plot during the time of experimenting. However, this plot received one treatment with a fertilizer, ProScape 32–0-6 (32% nitrogen, 6% soluble potash, 3% sulfur, and 6% chlorine) at 330 kg/ha in March 2009 and one treatment with the same fertilizer (at the same dose) in March 2010. Each trap was baited with 125 ml of 45.5% isopropanol (lure), either in a dispenser equipped with a wick of respective diameter or poured into the trap. The traps were hung in the open, on steel poles, at the height of 1–1.2 m. After 48 h the insects were collected and green June beetle adults identified according to Goodrich (1966) and the volume of the lure and percentage of isopropanol in the lure remaining in each trap or dispenser were measured. This experiment was replicated seven times in late July and early August 2010. On the basis of the experiment with emission rates we selected dispensers with 8 mm wicks for the remaining experiments.

Isopropanol Concentration. The experiments aiming to optimize isopropanol concentration in the lure were performed in Missouri State University peach orchards and in an apple/peach orchard near Berryville, AR. Improved bottle traps were baited with dispensers containing one of five concentrations of isopropanol 10, 33, 50, 66, or 91% and equipped with an 8 mm diameter wick protruding 8 mm above the bottle neck. Additionally, we used six control traps each baited with the commercially available green June beetle lure using 3.5 ml of the TRE8607 blend in a Trécé floral lure cup (Great Lakes Integrated Pest Management (IPM), Vestaburg, MI). Six blocks (replicates) along the mowed edge of grassy fields adjacent to apple or peach plantings were established near Berryville, AR. Following a Latin square design, each bottle trap at each randomly assigned location in each block dispensed one of six lures: one of five concentrations of isopropanol or the TRE8607 blend. Each trap was set at 1 m height on an aluminum stand and traps spaced 20 m apart. Traps were emptied daily and numbers of green June beetles per trap recorded. This experiment was performed in July 2010. During the time of the experiment, the plots in Berryville received one spray with an herbicide, Roundup (Monsanto Company, St. Louis, MO) at 2.4 liters/ha, and two sprays with insecticides: one with Actara 240 SC (thiamethoxam) at 100 ml/ha and one with Permethrin 3.2 EC (permethrin) at 740 ml/ha.

Additionally, perimeter apple trees surrounding the peach plots were baited with two grandisoic acid lures and with four benzaldehyde lures throughout the duration of the experiment (see Prokopy et al. 2004 for details about the dispensers). These trees received two sprays with Pyganic EC 5 (pyrethrines) applied at rate 1.184 liters/ha by a Solo backpack sprayer. Our traps were placed at a distance larger than 20 m from these trees. No fertilizer was used here in 2009 and 2010.

The plot in Mountain Grove received one spray with a fungicide, Topsin-M 70 WSB (thiophanate) at 1.73 kg/ha. Additionally, this plot received one spray with a fertilizer, ProScape 32–0-6 (32% nitrogen, 6% soluble potash, 3% sulfur, and 6% chlorine) at 330 kg/ha in March 2009 and one treatment with the same fertilizer (at the same dose) in March 2010.

In July and August 2011, another series of experiments was made in Mountain Grove, MO. On the basis of the experiments that optimized lure emission rates, only the dispensers with 8 mm cotton wicks were used. Five improved bottle traps, each equipped with the dispenser containing either 10, 33, 45.5, 66, or 91% aqueous isopropanol were randomly placed in the peach orchard at 1.0–1.3 m from the ground. The insects were collected after 24 h and identified according to Goodrich (1966). There were 17 replications of this experiment. Here, the plot received one spray with a fertilizer, ProScape 32–0-6 (32% nitrogen, 6% soluble potash, 3% sulfur, and 6% chlorine) at 330 kg/ha, in March 2010, and one spray with the same fertilizer (at the same rate) in March 2011. One spray with a fungicide, Abound (azoxystrobin) at 1.04 kg/ha was applied in July.

Optimal Location of the Traps in Relation to the Ground. To determine optimal heights at which the traps should be placed, a line of 15 metal fence posts was set up in July 2010 along the border of the aforementioned peach orchard in Mountain Grove, MO. The distance between the poles was equal 2 m. On each post, three potential trap heights: 0.5, 1.0, and 1.3 m were marked using black cable ties. Fifteen improved bottle traps, each baited with the dispenser containing 45.5% aqueous isopropanol and equipped with 8 mm wick were placed on the posts, one trap per post. The height to hang each trap was assigned randomly. The insects were collected after 48 h and identified according to Goodrich (1966). There were two trials in this experiment. The peach orchard received one spray with ProScape 32–0-6 (32% nitrogen, 6% soluble potash, 3% sulfur, and 6% chlorine) at 330 kg/ha, in March 2009 and one treatment with the same fertilizer (at the same dose) in March 2010. Additionally, a fungicide, Topsin-M 70 WSB (thiophanate) at 1.73 kg/ha was applied once during the time of the experiment.

In July 2010, a corresponding experiment performed in an apple/peach orchard near Berryville, AR, another Latin square design (four replicates), had improved bottle traps set 20 m apart at one of four treatment heights: 0.5, 1, 1.5, and 2 m. The traps were randomly assigned one of four sites in each of four blocks and each trap similarly baited with a dispenser filled with 125 ml of 91% isopropanol and equipped with 8 mm wick. Traps were emptied daily, numbers of green June beetles per trap recorded and traps were rerandomized according to the Latin square design. During the time of the experiment, the plots in Berryville received one spray with an herbicide, Roundup at 2.4 liters/ha, and two sprays with insecticides: one with Actara 240 SC (thiamethoxam) at 100 ml/ha and one with Permethrin 3.2 EC (permethrin) at 740 ml/ha.

Additionally, perimeter apple trees surrounding the peach plots were baited with two grandisoic acid lures and with four benzaldehyde lures as in Prokopy (2004) throughout the duration of the experiment. These trees received two sprays with Pyganic EC 5 (pyrethrines) applied at rate 1.184 liters/ha by a Solo backpack sprayer. Our traps were placed in a distance larger than 20 m from these trees. No fertilizer was used here in 2009 and 2010.

Trap Color. To determine if the color of the strip placed on the trap influenced the trap efficacy, nine improved bottle traps were prepared by stapling the top of each trap with a 4-cm wide transparent, red, orange, yellow, blue, black, purple, white, or green polypropylene band. The shade and color of each band was visually approximated to the closest matching color in Pantone solid color formula guide. The dominant wavelength, percentage reflected spectrum, and CIE $L^*a^*b^*$ values for each plastic color band was determined by a Jaz spectrometer (Ocean Optics, Dunedin, FL), where $L^* = 0$ (black) to 100 (white); $a^* =$ negative (green) to positive (red); and $b^* =$ negative (blue) to positive (yellow) (Table 1).

Each trap was baited with 100 ml of 45.5% isopropanol in a dispenser equipped with 8 mm wick. The traps were hung in a line on metal fence poles along the border of the aforementioned Missouri State University peach orchards in Mountain Grove, MO. The distances in between the poles equaled 2 m. The trap colors were randomly assigned to particular poles (one trap color per pole). The insects were collected after 24 h and identified according to Goodrich (1966). This experiment was repeated seven times in July and August 2010. The peach orchard received one spray with ProScape 32–0-6 (32% nitrogen, 6% soluble potash, 3% sulfur, and 6% chlorine) at 330 kg/ha, in March 2009, and one spray with the same fertilizer at the same dose in March 2010. One spray with a fungicide, Topsin-M 70 WSB (thiophanate) at 1.73 kg/ha was applied in July 2010. No sprays were applied in August 2010. On the basis of this experiment blue band was selected for following experiments.

Comparison of the Improved Trap with the Prototypical Standard Trap. Seven standard bottle traps (each with 125 ml of 45.5% isopropanol, topped with yellow band) and seven improved bottle traps (8 mm wick dispensers, 45.5% isopropanol) topped with blue band, were randomly placed in the commercial Catawba vineyard in Missouri State University experiment farm in Mountain Grove, MO. This experimental plot was located ≈50 m apart from the aforementioned peach orchard. The traps were hung at the height of 1.3 m above the ground level, 4–5 m apart. The insects were collected after 48 h and identified according to Goodrich (1966). This experiment was repeated twice in August, 2011. During the experiment, the plot was sprayed once with a fungicide, Elevate 50 WDG (fenhexamid) at 1.1 kg/ha. The plot also received two sprays with ProScape 32–0-6 (32% nitrogen, 6% soluble potash, 3% sulfur, and 6% chlorine) at 109.76 kg/ha, once in March 2010 and once in March 2011.

Statistical Analysis. Two treatment mean trap catches were compared by χ^2 test, whereas for data from three or more treatments we used analysis of variance (ANOVA) and means comparison by Bonferroni test or means separation by Waller–Duncan *k*-ratio *t*-test. For the trap catch for different color bands, the means separation was by Kruskal–Wallis test followed by Dunn's multiple comparisons of ranks. Results from experiments with emission rates were analyzed with ANOVA followed by Bonferroni comparison of means. Field efficacies of prototypical standard bottle trap with yellow band and improved bottle trap with a dispenser and blue band were compared with Student's *t*-test. χ^2 test, ANOVA with Bonferroni posttest, Kruskal–Wallis test followed by Dunn's multiple comparisons of ranks and Student's *t*-test were performed using GraphPad InStat version 3.00 (GraphPad Software, San Diego, CA). Remaining analyses used SAS (2008).

Results

Isopropanol Attractiveness. In total, 153 green June beetles were caught in standard bottle traps baited with standard isopropanol lure. No green June beetles were found in control traps containing aqueous 0.02% Triton. The results demonstrate that green June beetle are attracted to isopropanol in standard bottle traps ($P < 0.001$), not to the traps alone. Both sexes were attracted, indicating that isopropanol is not a sex-specific attractant.

Emission Rate. In a laboratory experiment, the isopropanol poured into the bottom of the standard bottle traps (without a dispenser) evaporated much faster than the isopropanol emitting from a bottle dispenser with a cotton wick hung inside the improved bottle traps. The volume of the isopropanol in standard bottle traps dropped from 125 ml to ≈60 ml within first 24 h of the experiment and to ≈35 ml after the next 24 h (Fig. 2A). Dispensers with the cotton wicks emitted isopropanol for much longer at steady rates ranging from 2.6 ml/d for a 2 mm diameter wick to 12.3 ml/d for 8 mm diameter wick (all wick surface areas

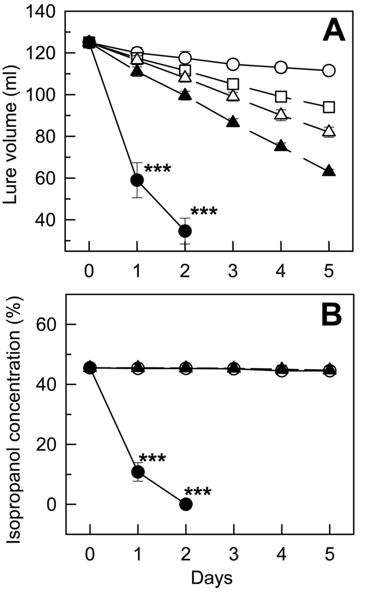

Fig. 2. Effect of wick diameter on isopropanol lure emission under laboratory conditions. (A) Time dependent changes in lure volume improved bottle traps equipped with dispensers with 2 mm wick (open circles, solid line), 4 mm wick (open squares, dashed line), 6 mm wick (open triangles dashed line), and 8 mm wick (solid triangles dashed line). Solid circles and solid line show changes in lure volume for standard open bottle trap. (B) Time dependent changes in concentration of isopropanol left in the lure in improved bottle traps equipped with dispensers with 2 mm wick (open circles, solid line) and 8 mm wick (solid triangles dashed line). Data for 4 and 6 mm wicks are omitted for clarity. Solid circles and solid line show changes in isopropanol concentrations for standard open bottle trap. Each data point represents mean ± SE ($N = 7$). $^{***}P < 0.001$ in Student's t-test.

were <3 cm^2) (Fig. 2A; $P < 0.001$). Dispensers with wicks also maintained a much more stable concentration of isopropanol than did the same isopropanol concentration with its much larger surface area (78 cm^2) exposed in the bottom of standard bottle traps. Throughout 5 d of the experiment, isopropanol concentration in the dispensers remained at the same stable level of 45–44.5%, whereas the concentration of isopropanol in the bottom of standard bottle traps dropped from 45% to ≈11% after 24 h and to 0.2% by 48 h (Fig. 2B; $P < 0.001$).

The corresponding field experiment showed that standard bottle traps were inferior to improved bottle traps both in terms of maintenance of constant isopropanol concentration, evaporation rate of isopropanol and capture of green June beetles (Table 2). Over a half of the isopropanol evaporated from standard bottle trap within 24 h leaving mostly water in the trap. Each standard bottle trap captured less than nine beetles per 48 h. Traps with cotton wick dispensers emitted lower amounts of the isopropanol per 48 h (from ≈9 to slightly above 43 ml per 48 h), but maintained amounts of emitted isopropanol at stable levels, which resulted in almost unchanged concentration of

Table 2. Dispenser wick diam effects on the vol of isopropanol solution emitted, percentage of isopropanol left in the dispenser, and avg no. of green June beetle ± SE ($N = 7$) caught per trap in Mountain Grove, MO, 2011

Wick diam (mm)	Isopropanol solution emitted within 48 h (ml)	% isopropanol left in solution after 48 h	No. GJB caught per trap within 48 h
None[a]	88.1 ± 6.5a	0a[b]	8.8 ± 1.2a
2	9.3 ± 1.8b	43.5 ± 1.5b	7.2 ± 1.4a
4	19.1 ± 2.3b	42.2 ± 1.7b	15.3 ± 1.9ab
6	26.5 ± 3.1c	42.0 ± 1.2b	27.1 ± 3.6c
8	43.3 ± 4.4c	43.2 ± 1.3b	57.2 ± 4.2d

The exp compared field performance of standard bottle traps with yellow bands (with 125 ml of 45.5% isopropanol, but without a dispenser or wick) and improved bottle traps with yellow bands and dispensers of 45.5% isopropanol equipped with cotton wicks of various diameters. Means followed by same letter do not differ significantly at $P > 0.05$ in ANOVA followed by Bonferroni comparison of means.

[a] Standard bottle trap without dispenser.

[b] Measured after 24 h.

this alcohol as it evaporated from the wick. Trap dispensers with cotton of 4, 6, or 8 mm in diameter caught significantly more green June beetles than standard bottle traps (Table 2; $P < 0.05$). On the basis of this experiment we selected dispensers with 8 mm wicks for the remaining experiments.

Isopropanol Concentration. In the experiment performed in Mountain Grove, MO, increased concentrations of isopropanol corresponded with increased numbers of green June beetles captured per trap. Concentrations of 45.5, 66, and 91% attracted more beetles than the lower isopropanol concentrations (Fig. 3; $P < 0.001$). No significant differences were found among any of the two medium concentrations tested (45.5 and 66%). However, 91% isopropanol attracted more green June beetles than 45.5% isopropanol ($P < 0.05$). A similar tendency was observed in the trials in Berryville, AR. There were no differences between beetle attractiveness of 50 and 66% isopropanol (Table 3) and in one case (the trial of 11 July) 99% isopropanol attracted significantly more beetles. However, in the remaining trials, 99% isopropanol was no more attractive to beetles than the dispensers con-

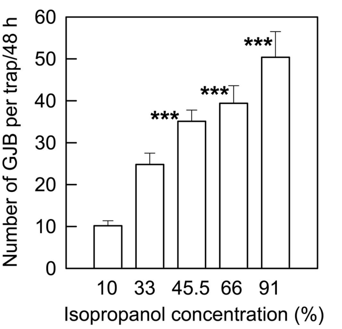

Fig. 3. Effect of isopropanol concentration on average number of green June beetles ± SE ($N = 17$) captured in the improved bottle traps with yellow bands in a peach orchard in Mountain Grove, MO, 2011. $^{***}P < 0.001$ in ANOVA followed by Bonferroni comparison of means.

Table 3. Average no. of green June beetles ± SE ($N = 6$) captured in improved bottle traps equipped with yellow bands and 8 mm cotton wicked dispensers containing various concentrations of isopropanol or baited with TRE8607 (the control)

Lure	No. green June beetles caught within 48 h		
	10 July	11 July	13 July
10% isopropanol	21.5 ± 5.1c	14.3 ± 6.1d	22.8 ± 4.9d
33% isopropanol	41.5 ± 9.0bc	51.7 ± 9.5c	64.0 ± 15.8c
50% isopropanol	77.3 ± 13.8ab	69.5 ± 14.0bc	89.2 ± 12.4bc
66% isopropanol	76.3 ± 11.9ab	77.0 ± 16.7b	105.2 ± 17.2ab
99% isopropanol	95.2 ± 28.9a	107.3 ± 9.8a	133.2 ± 24.7a
TRE8607[a]	46.8 ± 9.9bc	29.7 ± 28.9d	31.7 ± 5.2d

The exp was made in grass areas adjacent to apple or peach plantings near Berryville, AR, in July 2010. Means followed by same letter do not differ significantly at $P > 0.05$ in Waller–Duncan k-ratio t-test.

[a] 3.5 ml of TRE8607 blend dispensed twice weekly from a green Trící floral lure cup (Trící Inc., Adair, OK).

taining 50 or 66% isopropanol (Table 3; $P < 0.05$). In two trials on 11 July and 13 July, isopropanol at concentrations equal to or higher than 50% was more attractive to green June beetles than the TRE8607 lure (Table 3; $P < 0.05$).

Optimal Location of the Trap in Relation to the Ground. In Mountain Grove, MO, the traps hung at 1.3 m above the ground attracted significantly more beetles than the traps hung at lower heights (Fig. 4; $P < 0.01$). In Berryville, AR, the same tendency was observed (Table 4; $P < 0.05$). Additionally, it was found that elevating the traps to a height of 2 m did not significantly improve trap performance (Table 4).

Trap Color. The traps with blue, orange, and white bands attracted significantly more beetles than traps with transparent bands (Table 5; $P < 0.05$). Yellow, black, green, violet, and red bands were not as attractive (Table 5). Spectrometer analysis of the attractive bands (Table 1; Fig. 5) suggested that the beetles were attracted either to one of two light wavelength spectra; one covering the range between 450 and 475 nm (blue) and one corresponding to yellow and orange (570–620 nm).

Comparison of the Improved Trap with the Prototypical Standard Trap. The traps with blue bands and equipped with 8 mm wick dispensers of 45.5% isopropanol attracted about 10 times more green June beetles than the prototypical standard bottle traps with yellow bands (Fig. 6; $P < 0.001$).

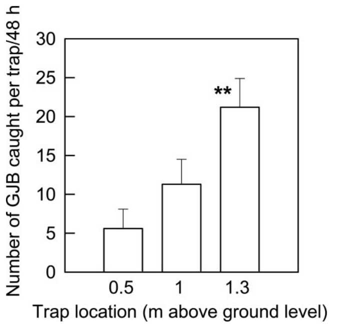

Fig. 4. Effect of trap location in relation to the ground on average number of green June beetles ± SE ($N = 10$) captured with 45% isopropanol baited improved bottle traps with yellow bands in Mountain Grove, MO, 2010. $^{**}P < 0.01$ in ANOVA followed by Bonferroni comparison of means.

Table 4. Effect of trap location in relation to the ground on avg no. of green June beetles ± SE ($N = 4$) caught in improved bottle traps equipped with yellow bands and 8 mm cotton wicked dispensers of 45.5% isopropanol near Berryville, AR, in July 2010

Trap ht (m)	No. green June beetles caught within 48 h		
	17 July	20 July	22 July
0.5	18.5 ± 4.8b	38.8 ± 4.4b	21.2 ± 2.5b
1	84.0 ± 15.6b	171.9 ± 52.0b	60.4 ± 18.4b
1.3	177.5 ± 50.8a	348.3 ± 44.5a	151.1 ± 21.5a
2	179.5 ± 24.2a	333.3 ± 67.5a	184.4 ± 68.6a

Means followed by the same letter do not differ significantly at $P > 0.05$ in Waller–Duncan k-ratio t-test.

Discussion

By combining the two ideas; 1) using easily obtainable isopropanol as the lure (Landolt 1990) and 2) using recycled polyethylene terephtalate beverage bottles for making the traps (Bambara et al. 2002) we have developed a simple and inexpensive trap for monitoring the green June beetle. The trap (Fig. 1e–k) has a detachable top (Fig. 1e), is equipped with an 8 mm cotton wick dispenser (Fig. 1h) that should dispense at least 125 ml of the lure per week. Field experiments with these dispensers emitted on average 43.3 ml of 45.5% isopropanol within 48 h. On extremely windy and hot days these dispensers emitted slightly >25 ml of 45.5% isopropanol per day (field experiments in Missouri, August 2011, data not shown). Such dispensers could be made of 125 Nalgene packaging bottles available from many vendors. Because, lures containing 45.5, 50, 66, 91, and 90% isopropanol exhibited similar attractiveness to green June beetles, we propose using 45.5% isopropanol as the most economic solution that could be obtained by mixing equal parts of isopropanol and water. In addition, the optimized trap for monitoring of green June beetles should be topped with a blue band. Water with surfac-

Table 5. Effect of band color (see spectrometer readings in Table 1) on avg no. of green June beetles ± SE ($N = 7$) caught in improved bottle traps equipped with 8 mm cotton wicked dispensers of 45.5% isopropanol in Mountain Grove, MO, in 2010

Color of the strap	Pantone color reference	No. green June beetles caught within 48 h
Clear		18.9 ± 3.1
Blue	2935C	68.7 ± 18.8**
Orange	165C	47.1 ± 7.9**
White	11–0602TPX	40.3 ± 4.6*
Yellow	109C	32.0 ± 4.2
Black	433C	31.3 ± 3.5
Green	354C	28.4 ± 5.8
Violet	226C	28.0 ± 2.4
Red	199C	19.2 ± 1.3

Means followed by stars ($^{*}P < 0.05$; $^{**}P < 0.01$) were significantly greater in comparison to traps with transparent straps (Kruskal–Wallis test followed by Dunn's multiple comparisons of ranks).

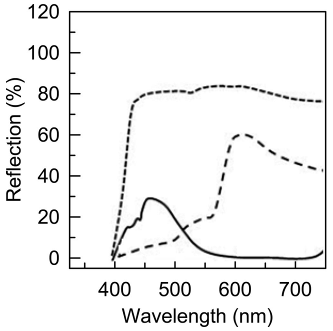

Fig. 5. Jaz Spectrometer (Ocean Optics, Dunedin, FL) measured optical spectra reflected by the polypropylene color bands that significantly ameliorated field attractiveness to green June beetles of improved bottle traps in Mountain Grove, MO, 2010; blue (solid line), orange (dashed line), and white (dotted line).

tant (125 ml) can be used as killing agent in the bottom of the bottle trap and hung at the height of 1.3 m above the ground. We successfully used scentless dishwashing liquid in proportion of one teaspoon per one gallon of tap water.

Our traps use the lure that is more attractive to the beetles than the only commercially available TRE8607 blend (Trécé Inc.). These traps are also reusable (we have used some traps for 3 yr in succession); they can be stored over winter in nonheated storage without loss of integrity. Our traps are inexpensive and are easy to procure; all components can be acquired from beverage bottle recycling centers or purchased in grocery stores or supermarkets. One trap could be made at a cost of less than \$7.20 the first season and \$4.70 the following season (Table 6). In the past, green June beetles were caught using baited JB Expando traps originally designed for the smaller Japanese beetle (*P. japonica*). That trap cost \$19 each and was the only commercial Japanese beetle trap with a large enough funnel opening to allow green June beetle adults to drop into the capture container.

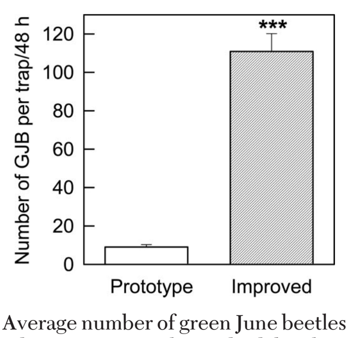

Fig. 6. Average number of green June beetles ± SE ($N = 14$) captured in prototypical standard bottle trap with a yellow band and baited with 125 ml of 45% isopropanol poured into the trap (open bar) versus that captured in the improved bottle trap blue band and 8 mm cotton wicked dispenser of 45% isopropanol (dashed bar). $^{***}P < 0.001$ in Student's *t*-test.

Table 6. Costs of our improved trap for monitoring the green June beetle, given in U.S. dollars according to 2011 retail prices and min. wages

Item	Season 1	Season 2	Season 3
Bottle[a]	0–0.98	None	None
Dispenser[b]	0.44–1.34	None	None
Lure[c]	3.36	3.36	3.36
Blue band[d]	0.02	0.02	0.02
Other materials[e]	0.25	0.07	0.07
Man-hour wages[f]	1.2	1.2	1.2
Total cost	5.27–7.15	4.65	4.65

[a] Sam's Choice soda bottle either recycled or purchased.
[b] Nalgene 125 ml packaging bottle and 12 cm of braided 8 mm clothesline for the wick.
[c] Lure of 45.5% isopropanol (a mix of rubbing alcohol, Cumberland Swan, Smyrna, TN, and water at V/V ratio of 1:1, 12 refills at \$0.28 each).
[d] Color band cut out from polypropylene report cover. Two 7-cm wide bands per season (the bands occasionally need replacement).
[e] Paper clips, staples, surfactant, 1 m of heavy duty polypropylene cord.
[f] Making the trap and its maintenance (mending, adjusting, etc.) at 10 min per trap at federal minimum wage. In the region that green June beetle range the minimum wages are equal to federal minimum wage or lower.

Our affordable monitoring trap could also be used in research on green June beetles where large numbers of traps need to be dispatched and maintained, for instance in studies of green June beetle biology, which still remains understudied. In particular, not much information has been reported since the early field reports by Davis and Luginbill (1921) and Chittenden and Fink (1922) in terms of phenology, habitat preference, dispersal, and population dynamics. We think that this could be, at least in major part, attributed to the fact that inexpensive monitoring tools were unavailable. Simplicity and low costs of our traps make it possible to continue research on aforementioned topics in variety of landscapes at minimal costs. We are currently working on spatial and temporal aspects of green June beetle distributions as well as a day-degree model for predicting adult emergence once a significant rain has moistened the soil enough to allow adults to dig to the surface.

Chromametric analysis of the trap color bands (Table 1; Fig. 5) also raises interest in some aspects of visual sensitivity of green June beetle adults. As far as visible spectrum of light is concerned, coleopterans were reported to be visually sensitive to violet (380–450 nm), green (500–557 nm), and orange (620 nm) (Briscoe and Chittka 2001). In our study, blue (450–475 nm), orange, and white were more attractive than yellow to green June beetle adults (Table 5). Interestingly, blue and white traps were reported as attractive to another scarab, Japanese beetle (Fleming et al. 1940). Ladd and Klein (1986) found that white and blue traps catch similar numbers of Japanese beetles and perform better than yellow traps. Yellow, green, red, and black traps attracted similar numbers of Japanese beetles in their study, a findings resembling our results for green June beetles. Another scarab, *Macrodactylus* *subspinosus*, preferred white traps to yellow traps (Williams et al. 1990). It cannot be excluded that preference of blue and white to yellow occurs in other scarabs too. More experimenting on color vision in *Scarabaeidae* beetles is needed.

Acknowledgments

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