Developing Monitoring Techniques for the Invasive Goldspotted Oak Borer (Coleoptera: Buprestidae) in California

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Environ. Entomol. 43(3): 729–743 (2014); DOI: http://dx.doi.org/10.1603/EN13162

ABSTRACT The goldspotted oak borer, *Agrilus auroguttatus* Schaeffer (Coleoptera: Buprestidae), is an invasive species that has colonized oak woodlands in southern California. To better define its seasonal flight activity, assist with forest and integrated pest management activities, and define the current distribution in California, an effective monitoring technique for *A. auroguttatus* is necessary. We assessed the efficacy of two colors of flight-intercept prism traps, the placement of these traps at three heights, and several commercially available lures [Manuka oil, Phoebe oil, and a green leaf volatile, (3Z)-hexenol] for monitoring the flight of adult *A. auroguttatus*. Landing rates and the densities of D-shaped emergence holes of *A. auroguttatus* adults were assessed on the lower stems of coast live oak, *Quercus agrifolia* Née, of varying size and crown health classes. Purple flight-intercept prism traps placed at heights of 3 m and 4.5 m caught significantly more female *A. auroguttatus* than green prism traps. In one experiment, males also responded at a significantly higher level to purple than to green prism traps placed at 3 m height. The addition of commercially available lures significantly enhanced male, but not female, *A. auroguttatus* trap catch when compared with unbaited control traps. There were no differences among male flight responses to the three lures. *A. auroguttatus* landing rates and emergence hole densities were significantly greater on the largest-diameter trees (>76.2 cm diameter at breast height) and on trees with severe crown thinning or complete crown collapse. The annual increment in emergence hole densities was also significantly greater on trees with severe crown thinning or complete crown collapse. In three trapping studies over multiple years in southern California, the adult flight period began as early as mid-May, peaked in mid-June to early July, and ended in early- to mid-September. To demonstrate the efficacy of the detection method for *A. auroguttatus* (unbaited purple traps at 3 m height), a delimitation survey conducted from 2009 to 2012 confirmed that the species was only present in San Diego Co., but that the distribution was expanding northward.

KEY WORDS *Agrilus auroguttatus*, flight-intercept prism trap, oak mortality, phloem/wood borer, semiochemical

The invasive goldspotted oak borer, *Agrilus auroguttatus* Schaeffer (Coleoptera: Buprestidae), is a recently recognized threat to forest health in southern California (Coleman and Seybold 2008 a,b). From 2002 to 2013, *A. auroguttatus* has caused elevated levels of tree mortality, killing nearly 25,000 oaks within an area of 212,784 ha in San Diego Co. (U.S. Department of Agriculture Forest Service, Forest Health Monitoring (USDA FS, FHM) 2013). Introduced populations of *A. auroguttatus* in California are believed to have originated from southeastern Arizona (Coleman et al. 2012b). In its native region, *A. auroguttatus* is not a significant ecological or economic threat to forest health (Coleman et al. 2012a). The native range of *A. auroguttatus* is hypothesized to coincide with the distributions of Emory oak, *Quercus emoryi* Torrey, and silverleaf oak, *Quercus hypoleucoides* A. Camus, in southeastern Arizona, southwestern New Mexico, and northern Mexico (Coleman and Seybold 2011). Although it was first collected outside of this native range in 2004 in San Diego Co. (Westcott 2005), the role of *A. auroguttatus* in oak mortality was not discovered until 2008 (Coleman and Seybold 2008b). At field sites in California, *A. auroguttatus* has been observed to feed as larvae on four native oak species, but injury and mortality have been most frequently observed in large-diameter [more than ≈46 cm diameter at breast height (DBH)] coast live oak, *Quercus agrifolia* Née, and California black oak, *Quercus kelloggii* Newb. (Coleman et al. 2012a).

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In California, *A. auroguttatus* kills healthy trees after several years of repeated larval feeding along the main stem and larger branches (Coleman et al. 2011). Since 2002, tree mortality associated with *A. auroguttatus* has spread radially within San Diego Co. to a distance of ≈50 km from its likely initial point of introduction near Guatay, CA. (Coleman et al. 2012a). The preferred novel hosts of *A. auroguttatus* are distributed from northern Baja California Norte, Mexico, to southern Oregon, representing a potentially large geographical distribution of invasion with increased risk for widespread tree mortality. Furthermore, the threat of moving *A. auroguttatus* populations in cut wood represents a significant concern for establishing satellite populations (Jones et al. 2013). In 2009, an isolated population of *A. auroguttatus* was detected in western San Diego Co. (Marian Bear Memorial Park, North Clairemont, CA; Coleman and Seybold 2011, Coleman et al. 2012a), and in 2012, an isolated population was detected in eastern Riverside Co. (Idyllwild, CA; Jones et al. 2013). The risk to oaks outside of the Pacific coast region remains largely unquantified.

The biology and life history of *A. auroguttatus* were unrecorded before 2008 (Coleman and Seybold 2008a,b). Therefore, developing an effective monitoring technique was necessary to facilitate research on its biology, to assist with forest and integrated pest management activities, and to better determine the current distribution in California. Early detection of *A. auroguttatus* populations increases the probability of conducting successful management (Flint et al. 2013), but can be difficult because of the insect's cryptic immature life stages and the slow rates of tree mortality. Preliminary *A. auroguttatus* trapping studies (Coleman and Seybold 2008b, Seybold et al. 2010) revealed that purple flight-intercept prism traps developed for the invasive emerald ash borer, *Agrilus planipennis* Fairmaire (Francese et al. 2008), were more effective at catching adults when compared with other trap types.

Other *Agrilus* sp. such as *Agrilus bilineatus* (Weber) (in the eastern United States) and *Agrilus biguttatus* (F.) (in Europe) colonize stressed or dying trees (Haack and Benjamin 1982, Katovich et al. 2000, Moraal and Hilszczański 2000), likely responding to volatiles emitted from the hosts. Bark sesquiterpenes and green leaf volatiles elicited responses from *A. planipennis* when the semiochemicals were placed on flight traps (de Groot et al. 2008, Crook and Mastro 2010). Research data from these congeners (especially *A. planipennis*) were used to design and initiate a project on the flight responses of *A. auroguttatus* to various traps and lures.

Our objectives were to assess the effectiveness of two colors of flight-intercept prism traps, the placement of prism traps at three heights, and the efficacy of several commercially available *A. planipennis* lures in eliciting flight responses from *A. auroguttatus.* To provide a basis for future work in chemical ecology, we also monitored the landing rates of *A. auroguttatus* adults on *Q. agrifolia* stems from trees with varying degrees of crown die back to assess the attractiveness of declining host trees. Emergence hole densities of *A. auroguttatus* adults were also recorded on stems of *Q. agrifolia* with varying degrees of crown die back to link oviposition and development to decline in host health and to compare changes in the inferred population density of *A. auroguttatus* over a 1-yr period. The flight periodicity of *A. auroguttatus* was determined from the combined results of all of the tests. Finally, the best currently available detection tool was used to define the distribution of *A. auroguttatus* in California through a delimitation survey.

Materials and Methods

Effect of Trap Color and Placement Height on Flight Response of *A. auroguttatus*. An experiment to evaluate the impact of trap color and placement height on the flight response of *A. auroguttatus* was conducted between February and September 2009 on the Descanso Ranger District of the Cleveland National Forest (CNF; San Diego Co., CA). At this general location, we selected sites with signs of *A. auroguttatus* infestation and associated tree mortality (Coleman et al. 2011, 2012a) that represented two elevational zones and two distinct forest stand types. At lower elevation sites (1,110 m), *Q. agrifolia* dominated open, riparian oak woodlands, whereas at higher elevation sites (1,800 m), *Q. kelloggii* co-dominated the forest canopy with Jeffrey pine, *Pinus jeffreyi* Balfour (nomenclature as in Price et al. 1998). A historical mean annual maximum temperature of 18.6°C, a mean minimum temperature of 4.6°C, and a mean annual rainfall of 90.2 cm were recorded in the area (Cuyamaca, CA; Western Regional Climate Center (WRCC) 2012).

Three experimental plots were established in each elevational zone based on elevated *A. auroguttatus* infestation rates (>50%) and elevated levels of oak mortality (Coleman et al. 2012a). We reasoned that high population densities of *A. auroguttatus* would be necessary for us to discern potential treatment effects in the experiment. The plots were located at three lower elevation sites—Descanso Fire Station (N 32.85663°, W −116.62276°), Noble Canyon Trailhead (N 32.84881°, W −116.52288°), and Pine Creek Trailhead (N 32.83660°, W −116.54280°)—and at three higher elevation sites—Wooded Hills Recreation Area (N 32.84961°, W −116.43242°), Camp Ole Fire Station (N 32.88107°, W −116.43045°), and Mt. Laguna (N 32.85642°, W −116.45511°). These plots were located within 20 km of each other.

At each of the six plots, three unbaited purple and unbaited green flight-intercept prism traps (three-sided, 35.6 by 59.7 cm, Francese et al. 2008) were hung in a vertical orientation so that the top of each trap was located at one of three heights (1.5, 3.0, and 4.5 m above the ground). Tangle-Trap Sticky Coating (The Tanglefoot Company, Grand Rapids, MI) was applied to the exterior surface of the traps to immobilize insects that landed on the trap surface. Traps were placed adjacent to the stems (i.e., within 3 m) of infested oaks to assist with the height arrangement and to reduce the variability in population sampling among the trap locations. Individual *Q. agrifolia* and *Q. kelloggii* were used to hang traps at the lower and higher elevation sites, respectively, and these trees were the focal point of each plot. All focal oaks were moderately to severely infested by *A. auroguttatus,* as determined by the degree of crown thinning, the density of emergence holes and bark staining, and the presence of signs of woodpecker foraging (Coleman et al. 2011, Hishinuma et al. 2011). Each trio of purple and green prism traps next to their focal tree was separated by 50 m at each plot location. Stainless steel conduit [e.g., Wheatland Electrical Metallic Tubing, Issue No. TT-548 (or IT-548), 1/2 in (1.3 cm) × 10 feet (3.05 m) EMT, UPC 8669202001] supported by a 3- to 4-foot (91.4–121.9 cm) rebar stake [1/2 in (1.3 cm) diameter] driven into the ground (Coleman and Seybold 2008b) held traps at 1.5 and 3.0 m, whereas rope tied to branches supported the trap at 4.5 m above the ground.

Traps were monitored weekly from 16 February to 9 September 2009 (29 wk) to assess flight periodicity of adult *A. auroguttatus;* only catches from 22 May to 9 September (i.e., 15 wk) yielded *A. auroguttatus.* Suspected adults were removed from the traps with forceps, transferred to vials or plastic bags, and returned to the laboratory for microscopic examination to confirm their species status and to determine their sex. Males were identified by the presence of the median groove on the first abdominal segment; this groove is absent on the females (Coleman and Seybold 2010).

Effect of Trap Color and Lure on Flight Response of *A. auroguttatus.* In 2009, we assessed the potential for cross-attractiveness of commercially available *A. planipennis* lures to adult *A. auroguttatus* by establishing three experimental plots in the lower elevational zone on the Descanso Ranger District of the CNF. At each plot, Pine Creek Trailhead (N 32.83660°, W −116.54280°), Buckman Springs Rd (N 32.80060°, W −116.50164°), and Guatay, CA (N 32.84919°, W −116.55044°), we placed one transect of four purple flight-intercept prism traps and one transect of four green flight-intercept prism traps. Traps within a transect were separated by >50 m and each transect was separated by >150 m within a plot. The tops of the traps were aligned evenly with the top of a stainless steel conduit (i.e., 3 m above the ground as described earlier) and each conduit pole was located adjacent (i.e., within 3 m) to the southern side of the crown of an *A. auroguttatus*-infested *Q. agrifolia* tree. The general levels of *A. auroguttatus* infestation and oak mortality in these plots were similar to those in the plots described earlier for the experiment on the effect of trap color and placement height on the flight response of *A. auroguttatus.*

The experiment was monitored from 5 June to 9 September 2009 (14 wk) and consisted of four semiochemical treatments associated with each of the two trap colors. The treatments were: 1) an unbaited trap, 2) Manuka oil (i.e., the steam distillate of the manuka tree (=New Zealand teatree), *Leptospermum scoparium* J.R.Forst. & G.Forst., product #3083 and 40 mg/d release rate of the crude distillate), 3) Phoebe oil (i.e., the steam distillate of the Brazilian walnut tree, *Ocotea* (=*Phoebe*) *porosa* (Nees & Martius) Barroso, product #P385-p and 40 mg/d release rate of the crude distillate), and 4) the green leaf volatile (3Z)-hexenol (product #3136 and 100 mg/d release rate; all products were purchased from Synergy Semiochemicals, Burnaby, BC, Canada; release rates established gravimetrically at 20°C by the vendor). Potential specimens of *A. auroguttatus* were removed from the traps weekly, at which time the treated traps were rerandomized among the four trap stations in each transect. Because there were three plots and 14 trapping periods, there were 42 trap catches per treatment for each of the two trap colors. The semiochemical lures were replaced once (mid-July).

Effect of *Q. agrifolia* Crown Condition on Landing Rate of *A. auroguttatus* on the Main Stem. In 2008 and 2009, we assessed the landing rate of *A. auroguttatus* adults on the main stems of *Q. agrifolia* at two experimental plots in the lower elevational zone on the Descanso Ranger District of the CNF [Scove Canyon (N 32.83056°, W −116.52003°) and Pine Creek Trailhead (N 32.83799°, W −116.54226°)]. The traps used in this experiment were clear plastic polyethylene panels (37.5 by 25 cm) that were coated with Tangle-Trap Sticky Coating and stapled in landscape orientation at the root collar (0 m), breast height (1.37 m), and 3.0 m above the ground on all trees. Trapping was initiated on eight *Q. agrifolia* on 17 July 2008 in Scove Canyon and on five *Q. agrifolia* on 15 May 2009 at Pine Creek Trailhead. Suspected adult *A. auroguttatus* were removed weekly until early September of each year and specimens were handled and evaluated as described earlier. The stem traps were placed in midseason in 2008 because *A. auroguttatus* and its damaging activity were first discovered in May and June 2008 (Coleman and Seybold 2008b). In 2009, panel traps were placed on *Q. agrifolia* with moderate to severe injury from *A. auroguttatus,* as determined by the degree of crown thinning (Coleman et al. 2011, Hishinuma et al. 2011).

In 2010 and 2011, we assessed the landing rate of adult *A. auroguttatus* across five crown health classes (Coleman et al. 2011) at three experimental plots in the lower elevational zone on the Descanso Ranger District of the CNF [Pine Creek Trailhead (N 32.83799°, W −116.54226°), Noble Canyon Trailhead (N 32.84879°, W −116.52341°), and Guatay, CA (N 32.84909°, W −116.55066°)]. Seventy-nine trees were monitored (40 trees in 2010; 39 trees in 2011), and each tree was considered a replicate. Traps were placed in landscape orientation at breast height on the northern and southern aspects of each tree monitored. Catches from the traps from the two aspects were summed to obtain one measurement per tree. The five crown health classes and numbers of trees represented in the experiment were: 1) healthy (no visible crown thinning, 18 trees), 2) light thinning (0–25% leaf loss, 22 trees), 3) moderate thinning (25–50% leaf loss, 21 trees), 4) severe thinning (>50% leaf loss, 13 trees), and 5) dead trees (no living foliage, 5 trees). Across all crown classes, the traps were placed on stems that spanned four diameter classes including <25.4 cm

(N = 2), 25.4–50.8 cm (N = 33), 50.8–76.2 cm (N = 26), and >76.2 cm (N = 18) cm DBH. Panel trap catches from the smallest size class (<25.4 cm DBH) were not included in the analysis owing to the small sample size for that group. Mean DBH of *Q. agrifolia* used in the study was 58.8 (±2.5 SE) cm. Trapping was initiated in late May in both years and suspected adult *A. auroguttatus* were removed once in early September when the study was concluded. Specimens were handled and evaluated as described earlier, except that the sex of the landing adults was not determined in 2010 and 2011 because there was no significant difference between the landing rates of the sexes in 2009 (see Results).

Correlation of *Q. agrifolia* Crown Condition With *A. auroguttatus* Emergence Hole Densities on the Main Stem. Adult *A. auroguttatus* make a distinctive D-shaped emergence hole through the bark surface of oaks (Hishinuma et al. 2011, Flint et al. 2013). *A. auroguttatus* is the only *Agrilus* sp. that attacks the main stem of *Quercus* species in California (Coleman and Seybold 2011, Coleman et al. 2011), and the only *Agrilus* sp. reared from the main stems of injured *Q. agrifolia* in San Diego Co. (Coleman and Seybold 2008b). In 2010 and 2011, the numbers of these holes were recorded from the stems of 146 *Q. agrifolia*, converted to a density, and correlated with the same five crown health class ratings of those trees as described previously. The primary analysis was conducted with *A. auroguttatus* emergence hole densities measured in 2011, but to compare the annual change in emergence hole densities across the crown classes, the densities were recorded in 2010 and 2011 from the same tagged trees. In 2011, trees in the following crown classes were assessed: 44 in class 1, 25 in class 2, 29 in class 3, 43 in class 4, and 5 in class 5 (146 trees total); each tree in each class was considered a replicate. Crown class rankings for the study trees changed little from 2010 to 2011. Sixty-five trees were surveyed in the 25.4–50.8-cm DBH size class, 52 trees in the 50.8–76.2-cm size class, and 26 trees in the >76.2-cm (largest) size class (143 trees total). Three trees had <25.4 cm DBH, and these were not included in the analyses. To determine the density of the D-shaped emergence holes, all holes were counted along the main stem from the root collar to 2.0 m in height. Counts were divided by the surface area of the respective 2-m stem section for each tree. Emergence holes were recorded between December and February during each year (i.e., while *A. auroguttatus* was in the larval or prepupal stage below the bark, Flint et al. 2013). Emergence holes were marked with an oil paint marker to facilitate an accurate accounting of the holes. Trees assessed with a negative change in emergence holes due either to miscounts or misidentification of the holes were not included in the analyses. These mistakes were rare, given the generally smooth bark surface of *Q. agrifolia.*

Application of Trapping Technique to Statewide Survey for *A. auroguttatus*. From 2009 to 2012, we used the unbaited purple flight-intercept prism traps to survey California for the presence or absence of *A. auroguttatus.* In this survey, traps were positioned so that the trap top was even with the top of the 3-m stainless steel conduit poles and were placed in the field from April until at least September during each year. This was a flexible and progressive survey whereby specific trap locations were varied with each year of the survey to maximize area-wide coverage of the state. Wherever possible, traps were placed adjacent to susceptible host trees and generally located at high-use sites (e.g., campgrounds, picnic areas, and trailheads). The survey was conducted primarily in the southern counties of the state (San Diego, Riverside, San Bernardino, and Los Angeles), but also included locations in El Dorado, Kern, San Luis Obispo, and Tulare Cos., which are central Sierra Nevada and southern Central Valley locations (Supp Data [online only]). Specimens were handled and evaluated as described earlier.

Statistical Analyses. All statistical analyses were conducted in SAS v. 9.2 (SAS Institute, Inc. 2010, Cary, NC, USA). A threshold of $\alpha = 0.05$ was used to determine significance in all tests. Data were untransformed, unless otherwise noted. Data from the experiment on the effect of trap color and placement height were analyzed by a generalized linear mixed model (PROC GLIMMIX), and the analysis was conducted separately by sex of *A. auroguttatus.* Sampling period, elevation (or oak species), trap color, trap placement height, and the interaction between trap color and trap placement height were the five fixed factors in the model. Site was nested in elevation and was considered as a random factor. The model used in the analysis was $\log[E(Y_{ijklm})] = \mu + T_i + E_j + C_l + H_m + (C{\times}H)_{lm} + S_k + \varepsilon_{ijklm}$, where Y_{ijklm} was *A. auroguttatus* trap catch during the i^{th} sampling period (i = 1–15); at the j^{th} elevation (j = low or high); at the k^{th} site (k = 1–6); for the l^{th} trap color (l = green or purple); and at the m^{th} placement height (m = low, middle, or high, i.e., 1.5, 3.0, or 4.5 m); μ was the overall mean. T_i, E_j, C_l, H_m, and $(C{\times}H)_{lm}$ denoted the fixed factors of sampling period, elevation, trap color, trap placement height, and the interaction between trap color and trap placement height, respectively. S_k denoted the random site effect assuming a normal distribution with zero mean and σ_s^2 variance. The residual error term (ε_{ijklm}) also assumed a normal distribution with zero mean and σ^2 variance. Trap catches were modeled conditionally on S_k as a negative binomial distribution, and *A. auroguttatus* trap catches were linked to their expected values with a log function. We assessed the extent of autocorrelation among sampling periods but concluded that it was weak because the model without covariance structure was similar to models with covariance structures such as unstructured, compound symmetry, or autoregressive (based on an analysis with the Akaike Information Criterion in SAS). We elected to address the issue of autocorrelation among sampling periods by using the first-order autoregressive method, which assumed a relationship among variances during the sampling periods, e.g., i_1 and i_2 as for two given sampling periods [type = ar(1) in SAS]. The Pearson's χ^2

statistic with 540 degrees of freedom (df) and the ratio of the χ^2 statistic to df were used to determine any dispersion in the trap catches. A P value >0.05 and a ratio close to one indicated no over- or under-dispersion of the data. The estimation method is maximum likelihood with adaptive quadrature (method = quad in SAS), and the method for computing the degrees of freedom of the denominator for the fixed effects was BETWITHIN (ddfm = bw in SAS).

Data from the experiment on the effect of trap color and lure were also analyzed by a generalized linear mixed model, and the analysis was conducted separately by sex of *A. auroguttatus.* The model used in this analysis was $\log[E(Y_{ijkl})] = \mu + T_i + C_k + L_l + (CL)_{kl} + S_j + \varepsilon_{ijkl}$, where Y_{ijkl} was *A. auroguttatus* trap catch during the i^{th} sampling period (i = 1–14); at the j^{th} site (j = 1–3); for the k^{th} trap color (k = green or purple); and to the l^{th} lure [l = 1–4, i.e., unbaited, Manuka, Phoebe, or (3Z)-hexenol]; μ was the overall mean. T_i was the sampling period effect, and was treated as a blocking factor because treatments were rerandomized among trap stations at the beginning of each sampling period. C_k, L_l, and $(CL)_{kl}$ denoted the fixed factors of trap color, lure type, and their interactions, respectively. S_j was the site effect, and it was treated as a random factor under the assumption of independence and a normal distribution with zero mean and σ_s^2 variance. The residual error term (ε_{ijkl}) also assumed a normal distribution with zero mean and σ^2 variance. Trap catches were modeled conditionally on S_j and with a negative binomial distribution, and *A. auroguttatus* trap catches were linked to their expected values with a log function. The analysis of autocorrelation among sampling periods, the estimation method, and the method for computing the degrees of freedom of the denominator for the fixed effects were the same as in the previous experiment. Multiple mean comparisons among lure types were conducted with least square means adjusted by Tukey's honestly significant difference test.

Catches of adult *A. auroguttatus* on stem panel traps (2008–2011) and emergence hole densities (2010–2011) were first evaluated for normality by using the Shapiro–Wilk test and for homogeneity of variances by comparing residuals (both PROC UNIVARIATE). Data from the two types of studies were $\log_{10}$ (catches +1) transformed to meet these assumptions; original (i.e., untransformed) data were presented in the figures. A general linear model (PROC GLM) was used to detect significant differences in adult *A. auroguttatus* trap catches for the landing rate (2008–2009) and emergence hole density (2010–2011) studies along the main stem. A mixed model (PROC MIXED) was used to detect significant differences in *A. auroguttatus* landing rates in 2010 and 2011. Means were separated with Tukey's honestly significant difference test if the null hypotheses of no significant difference among trap placement heights, DBH, or crown classes were rejected.

In the 2008–2009 landing rate study, data were analyzed with the model $Y_{ij} = \mu + X_i + \varepsilon_{ij}$, where Y_{ij} was the number of *A. auroguttatus* trapped on the j^{th} tree at the i^{th} trap height (i = 1–3); Xi was the i^{th} trap height; and ε_{ij} was the residual error. In the 2010–2011 landing rate study, data were analyzed to determine if there were any significant differences by DBH or crown class. These landing rates were analyzed by using a mixed model with year as a random factor (PROC MIXED). The model was $Y_{ijk} = \mu + X_i + Z_i + \varepsilon_{ijk}$, where Y_{ijk} was the number of *A. auroguttatus* trapped on the k^{th} tree in the i^{th} DBH (i = 1–3) or crown (i = 1–5) class in the j^{th} year (j = 1 or 2); μ is the overall mean; X_i was either the i^{th} DBH or crown class; Z_i was the error term associated with the random factor year; and ε_{ijk} was the residual error.

In the 2010–2011 emergence hole density study, emergence hole density in 2011 and the increment in emergence hole density from 2010 to 2011 were analyzed with the model $Y_{ij} = \mu + X_i + \varepsilon_{ij}$, where Y_{ij} was the *A. auroguttatus* emergence hole density in 2011 (or its annual increment) of the j^{th} tree in the i^{th} DBH (i = 1–3) or ith crown (i = 1–5) class; Xi was either the i^{th} DBH or crown class; and ε_{ij} was the residual error.

Results

Effect of Trap Color and Placement Height on Flight Response of *A. auroguttatus*. In total, 448 adult *A. auroguttatus* were collected in 2009 during this study (152 adults at the low elevation site and 296 at the high elevation site). There was a significant effect of sampling period on trap catch of *A. auroguttatus,* regardless of sex (male: $F_{14,70} = 8.97$, $P < 0.001$; female: $F_{14,70} = 9.59$, $P < 0.001$). Elevation (or oak species) did not affect trap catch of *A. auroguttatus,* regardless of sex (male: $F_{1,4} = 0.05$, $P = 0.83$; female: $F_{1,4} = 2.74$, $P = 0.17$). Interaction between trap color and trap placement height did not affect *A. auroguttatus* trap catch, regardless of sex (male: $F_{2,10} = 2.07$, $P = 0.18$; female: $F_{2,10} = 1.41$, $P = 0.29$). There was no significant effect of trap color on trap catch of male *A. auroguttatus* ($F_{1,5} = 0.55$, $P = 0.49$), but purple prism flight-intercept traps caught significantly more female *A. auroguttatus* than did green prism traps (Fig. 1A; $F_{1,5} = 15.05$, $P = 0.01$). There was no significant effect of height of trap placement on trap catch of male *A. auroguttatus* (Fig. 1B; $F_{2,10} = 0.05$, $P = 0.95$), but there was a significant effect on trap catch of females; more were caught when traps were hung 4.5 m above the ground when compared with traps hung at 1.5 m (Fig. 1B; $F_{2,10} = 4.55$, $P = 0.04$).

Although traps were monitored weekly from February to September, no *A. auroguttatus* adults were caught from February to mid-May. Female trap catch began during the week of 29 May to 4 June 2009 at both lower and higher elevation sites (Fig. 1C). The first males of the season were caught earlier (the week of 15–22 May 2009) than the first females at lower elevation sites and during the same period as females at higher elevation sites (Fig. 1C). Peak flight activity occurred from 25 June to 2 July for both sexes at the lower and higher elevation sites (Fig. 1C). The majority of *A. auroguttatus* (86%) were trapped from 25 June to 27 July across all sites, but catches occurred into early September.

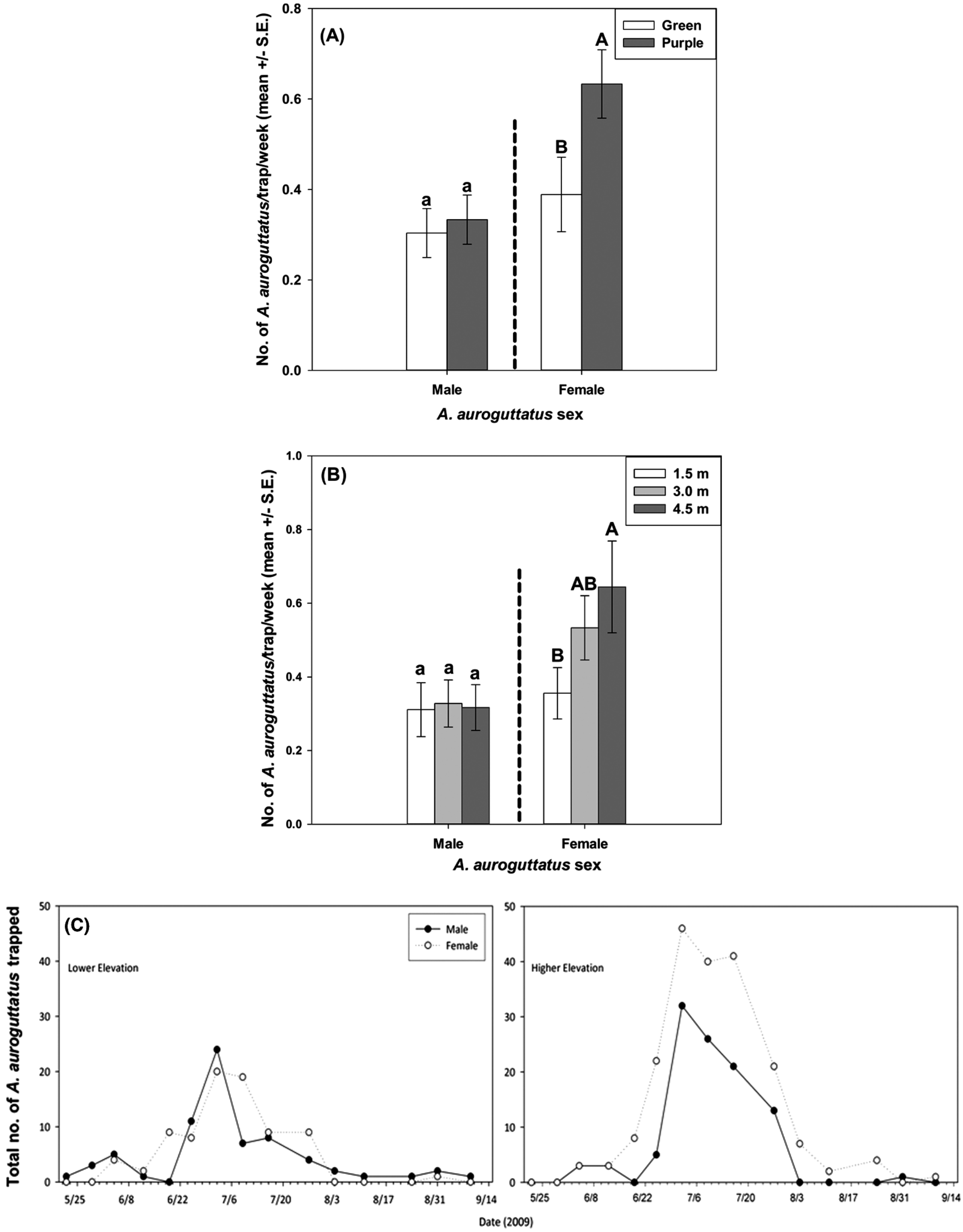
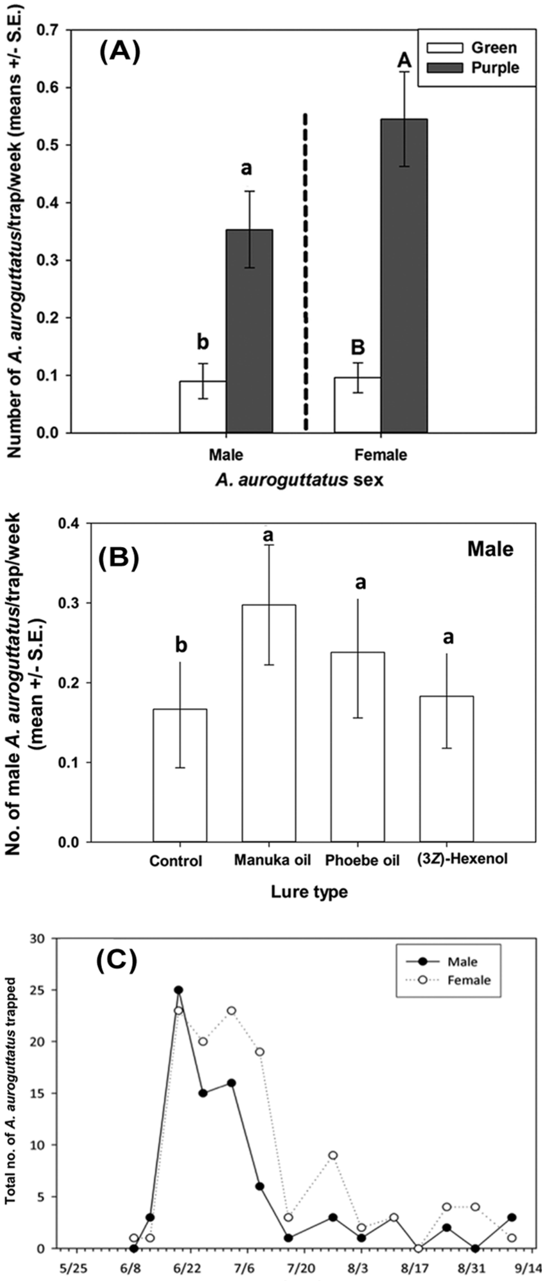


Fig. 1. Mean number (±SE) of male and female adult goldspotted oak borers, *Agrilus auroguttatus*, trapped in 2009 with unbaited purple and green flight-intercept prism traps (A) at three heights (B) and the corresponding periodicity of the flight response to these traps at lower and higher elevation sites (C) in San Diego Co., CA. Different lower- or uppercase letters above the means of male or female *A. auroguttatus* catches, respectively, denote a significant difference between trap colors (A, female: $F_{1,5} = 15.05$, $P = 0.01$) or among trap heights (B, female: $F_{2,10} = 4.55$, $P = 0.04$). Means in panels A and B were calculated based on sample sizes of $N = 270$ and $N = 180$, respectively. At the higher elevation site (C), male and female flight was identical during the first four assay points.

Effect of Trap Color and Lure on Flight Response of *A. auroguttatus*. In total, 191 *A. auroguttatus* adults were collected in 2009. There was a significant effect of sampling period on trap catch of *A. auroguttatus*, regardless of sex (male: $F_{13,26} = 4.23$, $P < 0.001$; female: $F_{13,26} = 4.90$, $P < 0.001$). The interaction between trap color and lure type was not significant for either sex (male: $F_{2,6} = 1.15$, $P > 0.05$; female: $F_{2,6} = 1.15$, $P > 0.05$). There were significant effects of trap color and lure type on trap catch of male *A. auroguttatus* (Fig. 2A, color: $F_{1,2} = 352.49$, $P = 0.002$; Fig. 2B, lure: $F_{3,6} = 219.42$, $P < 0.001$); purple traps were more attractive than green traps (Fig. 2A). All three baited trap treatments were more attractive to male *A. auroguttatus* than unbaited traps; however, there were no differences in responses among the lure treatments (Fig. 2B). There was also a significant effect of trap color on trap catch of female *A. auroguttatus* (Fig. 2A; $F_{1,2} = 37.70$, $P = 0.03$), but there was no effect of lure type on trap catch of females ($F_{1,2} = 0.37$, $P > 0.05$).



Initial catches of *A. auroguttatus* began the week of 8–12 June 2009 for males and the week of 5–8 June 2009 for females (i.e., essentially as soon as the traps were placed in the field; Fig. 2C). For both sexes, beetles were trapped continuously until 17 August 2009 (Fig. 2C). Peak *A. auroguttatus* trap catches occurred during the week of 12–19 June for males and females. Female trap catch peaked again during the week of 24 June to 2 July 2009. The majority of *A. auroguttatus* (87%) were trapped from 19 June to 27 July, but catches occurred until 9 September 2009.

Effect of *Q. agrifolia* Crown Condition on Landing Rate of *A. auroguttatus* on the Main Stem. Adult *A. auroguttatus* trap catch was highest numerically in 2008 and 2009 on the 3-m-high stem panel traps (Fig. 3A), but there was no significant difference in trap catch among panel trap heights. Eighty-six adults were collected in 2008, whereas 274 adults were collected in 2009. In 2009, mean (±SE) male [7.6 (±2.39), 114 total] and female [10.6 (±3.27), 160 total] trap catches were similar. Males and females were trapped from 1 July to 18 August in 2008 and from 15 May to 9 September in 2009 (Fig. 3B). Peak flight activity in 2009 occurred during the week of 25 June to 2 July. The majority of *A. auroguttatus* (85%) were trapped from 19 June to 27 July, but catches occurred until 9 September 2009.

In landing rate studies in 2010–2011, diameter class affected landing rates ($F_{2,73} = 6.13$, $P = 0.004$). Significantly more *A. auroguttatus* adults were trapped on stem panel traps placed at breast height on *Q. agrifolia* in the two largest-diameter classes (>76.2 cm DBH and 50.8–76.2 cm DBH) than in the smallest-diameter

Fig. 2. Mean number (±SE) of male and female adult goldspotted oak borers, *A. auroguttatus*, trapped in 2009 with purple and green flight-intercept prism traps (A), male *A. auroguttatus* trapped in 2009 in response to unbaited control traps and three different lure types (B), and the corresponding periodicity of the flight response to these traps (C) at lower elevation sites in San Diego Co., CA. Different lower- or uppercase letters above the means of male or female *A. auroguttatus* catches (A), respectively, denote a significant difference in trap catch between trap colors (male: $F_{1,2} = 235.49$, $P = 0.002$; female: $F_{1,2} = 37.70$, $P = 0.03$). Different lowercase letters above the means of male *A. auroguttatus* catches (B) denote significant difference among treatments. Means in panel A were calculated based on sample sizes of $N = 167$ (one trap catch was lost for each color of trap because both (3Z)-hexenol lures were lost from one plot between 8 and 12 June) and means in panel B were calculated based on sample sizes of either $N = 84$ (Manuka oil, Phoebe oil, and Control) or $N = 82$ [(3Z)-hexenol].

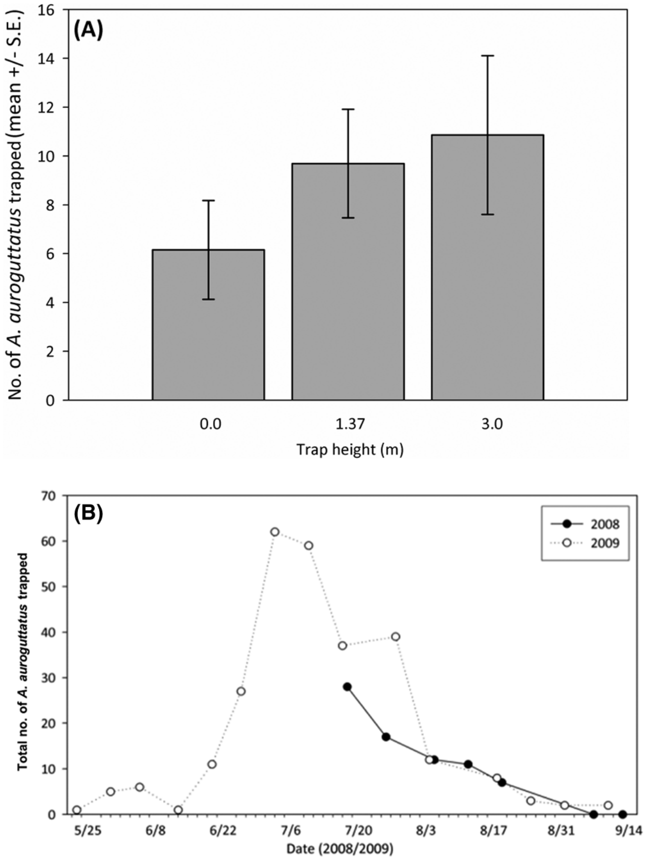


Fig. 3. Mean number (±SE) of adult goldspotted oak borers, *A. auroguttatus*, trapped in 2008 and 2009 (A) and the corresponding periodicity of the flight response (B) to clear panel stem traps placed at three heights on coast live oak, *Quercus agrifolia*, in San Diego Co., CA. Means in panel A were calculated based on sample sizes of $N = 13$, the number of trees in the study.

class (25.4–50.8 cm DBH; Fig. 4A). Crown class also affected landing rates ($F_{4,71} = 6.51$, $P < 0.001$). Adult *A. auroguttatus* landing rates were significantly lower on living oaks with no crown thinning than on oaks with severe crown loss; trap catch increased numerically but not significantly along a gradient among the lightly injured trees in the lower crown classes (crown classes of 2–3, respectively; Fig. 4B). There were no significant differences in landing rates (or flight and bark surface activity) of *A. auroguttatus* adults between dead trees (crown class 5) or trees with severe crown thinning (crown class 4); however, trap catches were greater on dead *Q. agrifolia* than on healthy trees with no apparent crown thinning (i.e., crown class 1; Fig. 4B).

Correlation of *Q. agrifolia* Crown Condition With *A. auroguttatus* Emergence Hole Densities on the Main Stem. In 2011, there was a significantly greater density of D-shaped *A. auroguttatus* emergence holes on trees in the largest-diameter class (Fig. 5A) and on trees with more severe crown health class ratings (Fig. 5B). Trees in the smallest size class (25.4–50.8 cm DBH) had an 82% lower emergence hole density than trees in the largest size class (>76.2 cm DBH). Trees with a crown rating of 5 (i.e., dead with no foliage) had a significantly greater emergence hole density than trees in other crown classes (Fig. 5B). The density of emergence holes was also significantly different on live trees with severe crown thinning (crown class of 4) when compared with live trees in the first three crown classes (1–3). No significant interactions were detected for density of *A. auroguttatus* emergence holes with diameter class or crown class ($F_{8,129} = 0.38$, $P = 0.9$).

There were positive changes in the numbers of D-shaped emergence holes in *Q. agrifolia* from 2010 to 2011 for each diameter class (Fig. 5C). The change in the density of D-shaped emergence holes from 2010 to 2011 was greatest in the largest-diameter class (>76.2 cm DBH) when compared with the two smaller-diameter classes, but this change was not statistically significant ($F_{2,141} = 0.54$, $P = 0.585$, Fig. 5C). There was a significant effect of crown class on the change in the density of D-shaped emergence holes (Fig. 5D). This change was greatest on trees that died within the past year (crown class of 5, Fig. 5D) and significantly different from the first four crown classes (1–4). The change in density of D-shaped emergence holes on live trees with severe crown thinning (crown class of

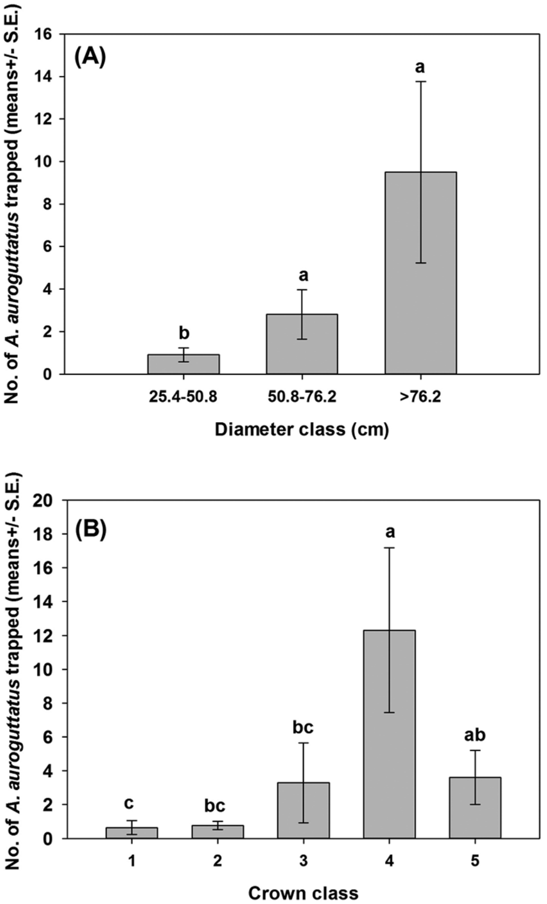


Fig. 4. Mean number (±SE) of adult goldspotted oak borers, *A. auroguttatus*, trapped in 2010 and 2011 across three diameter at breast height (DBH) classes (A) and five crown classes (B) with clear stem panel traps placed on coast live oak, *Q. agrifolia*, in San Diego Co., CA. Different lowercase letters above histogram bars within a figure panel indicate significant differences among means (panel A: $F_{2,73} = 6.13$, $P = 0.004$; panel B: $F_{4,71} = 6.51$, $P < 0.001$). Means in panel A were calculated (left to right) based on sample sizes of N = 33, 26, and 18; means in panel B were calculated (left to right) based on sample sizes of N = 17, 21, 21, 13, and 5.

4) was significantly different when compared with live trees in the first three crown classes (1–3; Fig. 5D). No significant interactions were detected for the annual increment in the density of *A. auroguttatus* emergence holes with diameter (DBH) or crown class ($F_{8,129}$ = 1.08, $P = 0.38$).

Application of Trapping Technique to Statewide Survey for *A. auroguttatus*. In total, 161 purple prism flight traps were monitored from 2009 to 2012 across 132 sites in California (Fig. 6A and Supp Data [online only]). During this period, *A. auroguttatus* was detected on 51 traps in San Diego Co. (Fig. 6B). In 2012, traps detected *A. auroguttatus* populations on the northern edge of the infestation in San Diego Co. where no evidence of tree injury or mortality had been observed. The area delimited by positive trap catches covered ≈300 km^2.

Discussion

Tree mortality associated with *A. auroguttatus* represents the most significant insect threat to oak woodlands in southern California. As *A. auroguttatus* populations continue to spread naturally or via human-assisted dispersal in firewood, early detection of populations will be crucial for initiating integrated pest management techniques at high-value sites. Development of an effective trap, lure, and survey protocol can facilitate the implementation of a successful integrated pest management program.

Sticky panel flight-intercept traps have been effective for trapping various buprestid species, including *A. bilineatus* and *A. planipennis* (Montgomery and Wargo 1983; Francese et al. 2008; Crook et al. 2009; McCullough et al. 2009, 2011). Purple flight-intercept prism traps were more effective at collecting *A. auroguttatus* females than green flight-intercept prism traps in two of our studies and were more effective at collecting males in one of our studies. Catches of female *A. auroguttatus* were numerically higher when traps were hung at 3 and 4.5 m above the ground and significantly higher at 4.5 m. Female *A. planipennis* were also collected more frequently on purple prism traps, whereas male *A. planipennis* preferred green prism traps when the traps were hung in the midcanopy of host trees (Francese et al. 2008, Crook et al. 2009). Despite the preference for colonizing the basal portion of the tree (Coleman et al. 2011, Haavik et al. 2012), hanging traps higher in the canopy or implementing a double-decker trapping arrangement (Poland et al. 2011) may be more efficient for collecting *A. auroguttatus* adults. The adults are likely associated with foliage in the crowns of oaks as they feed and mate (Flint et al. 2013).

Our recommendation for trapping *A. auroguttatus* is to suspend unbaited, sticky purple flight-intercept prism traps on the top of 3-m metal conduit poles and place them adjacent to a host species on the edge of a stand or in direct sunlight. Components of the volatiles found in commercially available *A. planipennis* lures are present in oaks (Palma-Fleming and Kepner 1983, Christoph et al. 1999). However, these lures did not elicit a significant increase in flight response by female *A. auroguttatus*, and significantly enhanced male response only at the level of about one-tenth of a beetle/wk (Fig. 2B). Thus, there is a questionable cost-benefit effect of using any of these lures in a detection or monitoring program for *A. auroguttatus.* Manuka and Phoebe oils, which contain high concentrations of antennally active ash volatiles, were more effective at increasing trap catches of *A. planipennis* on similar traps (Crook et al. 2008, Marshall et al. 2009). Gas chromatographic-electroantennographic detection assays of *A. auroguttatus* in response to oak mono- and sesquiterpenes will be required for the development of an effective lure. Ethanol has been an effective semiochemical lure for other wood borers, bark beetles, and

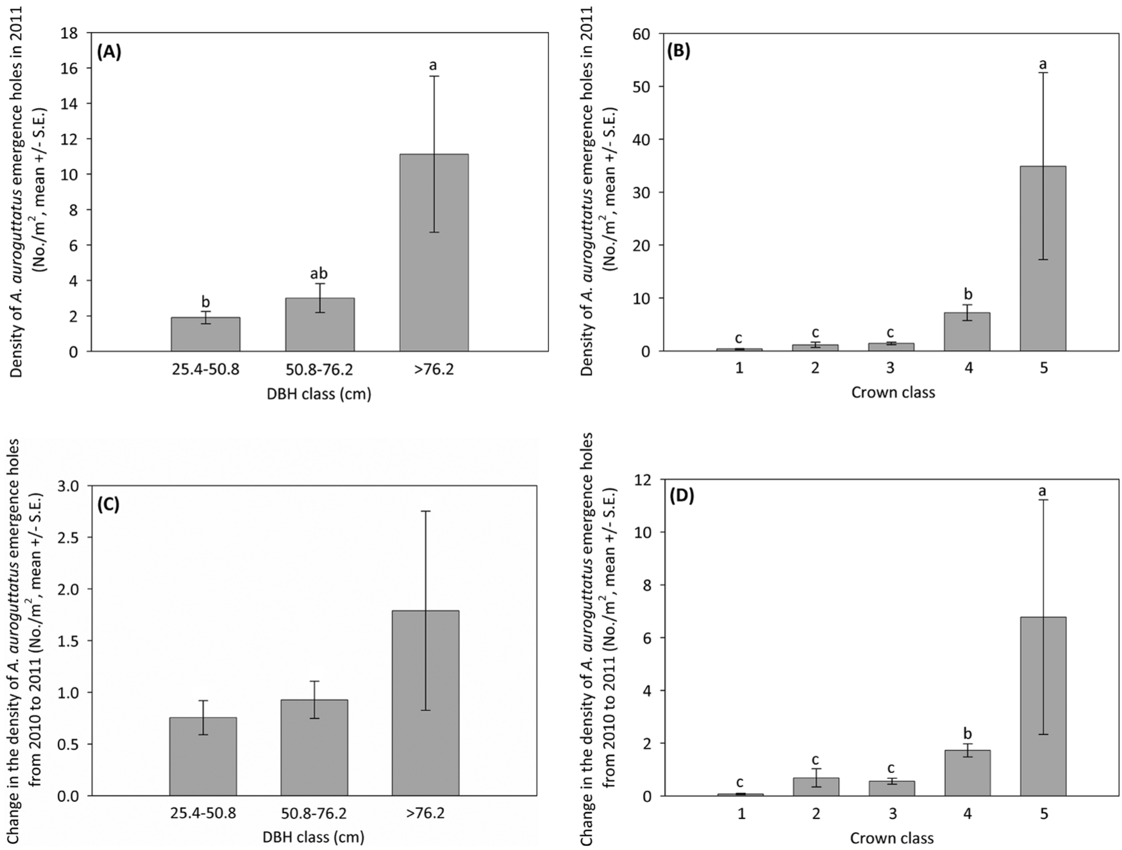


Fig. 5. Density (No./m^2, mean ± SE) of D-shaped emergence holes from the goldspotted oak borer, *A. auroguttatus*, recorded in 2011 across three coast live oak, *Q. agrifolia*, DBH classes (A) and five crown classes (B). The annual increment in the density of *A. auroguttatus* emergence holes (No./m^2, mean ± SE) from 2010 to 2011 was also assessed across three *Q. agrifolia* diameter classes (C) and five *Q. agrifolia* crown classes (D). Different lowercase letters above histogram bars within a figure panel indicate significant differences among means (panel A: $F_{2,141} = 3.26$, $P = 0.041$; panel B: $F_{4,139} = 37.93$, $P < 0.001$; panel C: $F_{2,141} = 0.54$, $P = 0.585$; and panel D: $F_{4,139} = 23.59$, $P = 0.001$). Means in panels in panels A and C were calculated (left to right) based on sample sizes of $N = 65$, 52, and 26; means in panels B and D were calculated (left to right) based on sample sizes of $N = 44$, 25, 29, 43, and 5.

ambrosia beetles (Montgomery and Wargo 1983, Miller and Rabaglia 2009, Kelsey et al. 2013), but preliminary trap catch data with *A. auroguttatus* from ethanol-baited traps have not been encouraging (Coleman and Seybold 2008b, Coleman, unpublished data). Ethanol may enhance *A. auroguttatus* trap catch when combined in a lure with other volatiles, as has been observed with *A. planipennis* (McCullough et al. 2011), so this should be investigated with *A. auroguttatus.* The addition of a green leaf volatile [(3Z)-hexenol] alone did not enhance trap catch of female *A. auroguttatus* to purple- or green-colored traps, but may have been more effective if traps were suspended closer to the crown of oaks or if this potential semiochemical were combined with other behaviorally active compounds.

Trap catches of *A. auroguttatus* adults on the purple and green flight-intercept prism traps were relatively low even in sites with high population densities, as inferred from emergence hole densities and host foliage loss. Thus, the use of the current trapping protocol for early detection of *A. auroguttatus* populations may have limited efficacy. However, purple flight-intercept prism traps detected *A. auroguttatus* populations in sites with no detectable tree injury and mortality in northern San Diego Co. (Fig. 6B). Until additional research reveals olfactory or visual (see later text) stimuli to improve the efficacy of the purple trap, our best tool for trapping *A. auroguttatus* is the unbaited sticky purple flight-intercept prism trap.

We interpret the trap catches of *A. auroguttatus* on the stem panel traps as a measure of adult landing rate, although we realize that some adults captured on these traps could in theory be newly emerged adults from the same stem on which the traps were placed. The synchrony of the trap catches in 2009 on purple prism traps (Figs. 1C and 2C) when compared with trap catches on the stem panel traps (Fig. 3B) suggests that the stem panel traps did indeed measure landing rate of flying adults dispersing from other trees. These rates for the females can be inferred as an indirect measure of oviposition. In 2008 and 2009, landing rates

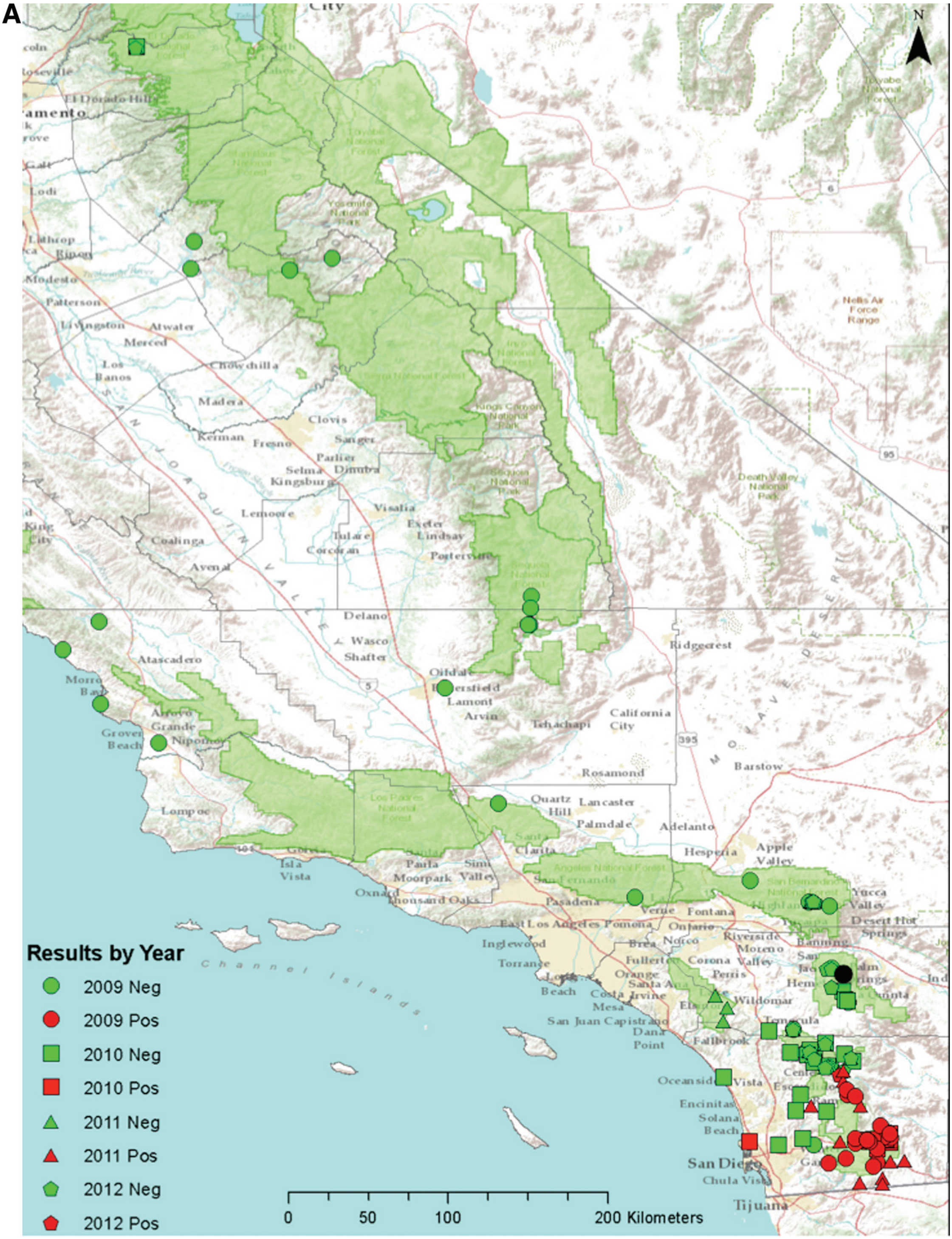


Fig. 6. Delimitation survey of California (A) and southern California (expanded view) (B) for the goldspotted oak borer, *A. auroguttatus*, indicating positive (red) and negative (green) trap catches from 2009 to 2012. The survey locations ranged from El Dorado Co. in the north (central Sierra Nevada) to San Diego Co. in the south. The black circle in both panels indicates the positive *A. auroguttatus* find in 2012 in Riverside Co. Public lands are indicated by green shading.

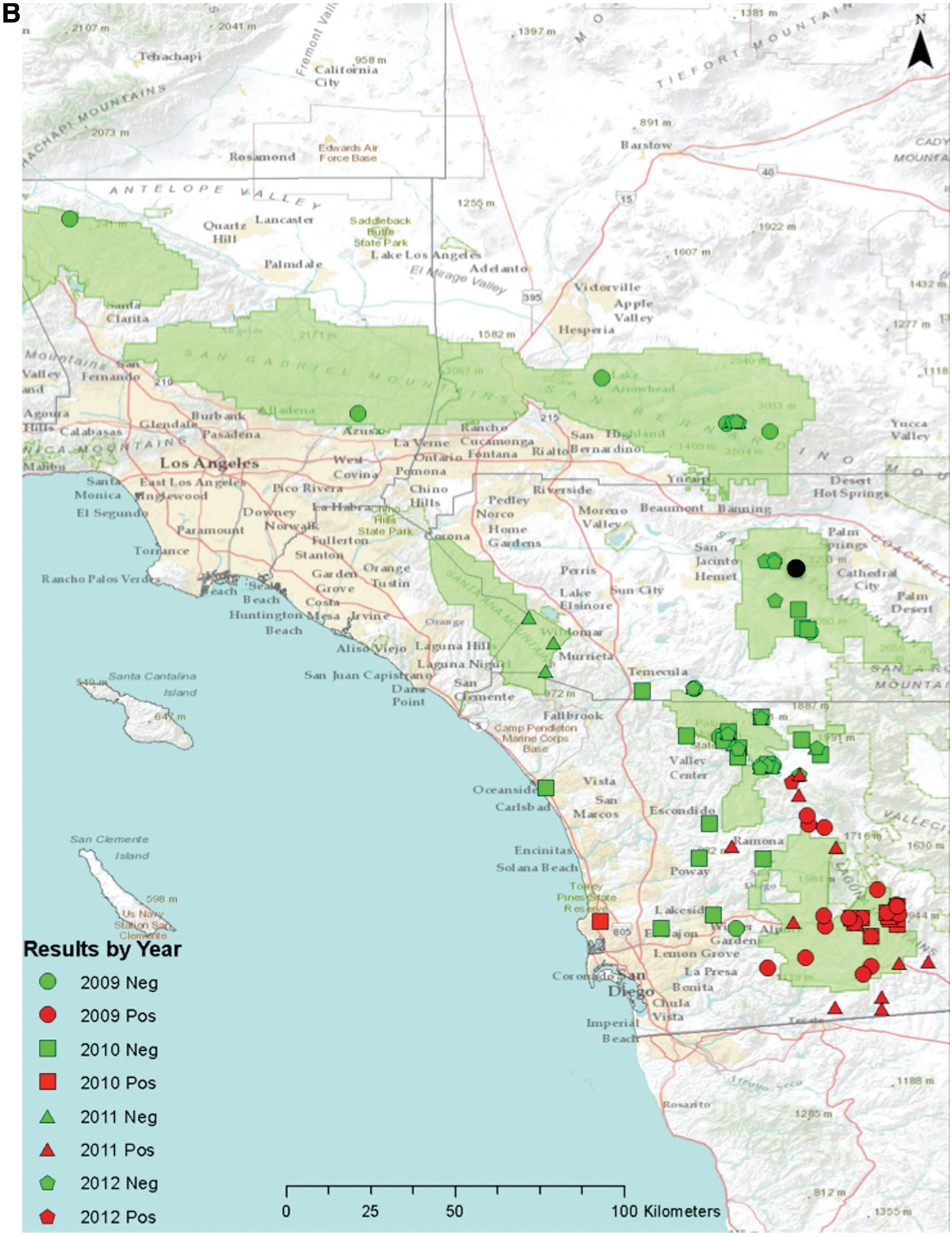


Fig. 6. (Continued).

of *A. auroguttatus* adults on *Q. agrifolia* did not vary across three heights along the lower portion (<3.0 m) of the main stem (Fig. 3A). Haavik et al. (2012) reported that emergence hole densities, which reflect host selection and oviposition by adult females, and subsequent injury from *A. auroguttatus* larvae (all behavioral events subsequent to landing), were randomly dispersed on the lower portion (<1.52 m) of the main stem. The similarity in male and female landing rates measured in 2009 on the stems of *Q. agrifolia* raises the question of why males would seek the main stem of the host at this stage in the life cycle and biology of *A. auroguttatus.*

In 2010 and 2011, stem panel trapping studies revealed that *A. auroguttatus* landing rates were significantly greater on *Q. agrifolia* stems in the largest-diameter class (Fig. 4A). Injury and mortality of *Q. agrifolia* and *Q. kelloggii* caused by *A. auroguttatus* in California have been observed predominantly on larger-diameter trees (>46 cm DBH; Coleman et al. 2012a), corresponding to the two larger-diameter classes (50.8–76.2 and >76.2 cm DBH) monitored in this study. These large-diameter trees appear to be at greatest risk for *A. auroguttatus*-caused tree mortality. As *A. auroguttatus* populations expand in California, ground surveys for early detection should focus on these large-diameter trees with declining crowns. Conversely, small-diameter trees may not be at significant risk for *A. auroguttatus*-caused mortality, as supported by the low landing rates recorded in our study and low levels of infested small-diameter *Q. agrifolia* in a previous survey (Coleman et al. 2012a).

In 2010 and 2011, *A. auroguttatus* landing rates were higher on oaks that had higher crown health classification ratings (an indirect measure of larval injury; Fig. 4B). However, the landing rates were lower on dead trees (crown class 5; Fig. 4B). This increase in attraction to more injured trees may represent a source of host- or insect-based semiochemicals that elicit flight responses from adult *A. auroguttatus.* The trend of elevated landing rates was also detected in 2009 when stem panel traps were placed on severely injured trees (Fig. 3A and B). Volatiles emitted from moderately and severely injured trees may be attractive to adults, emphasizing the need for collection of volatiles from various portions of attacked oak trees followed by gas chromatographic-electroantennographic detection studies. Volatiles of unknown chemical composition from the inner bark of stressed white oaks were attractive to *A. bilineatus* in various flight traps (Dunn et al. 1986a). The bronze birch borer, *Agrilus anxius* Gory, *A. bilineatus,* and *A. planipennis* have also responded in flight to stressed and girdled trees (i.e., severing a strip of phloem and/or xylem from a living tree; Anderson 1944, Dunn et al. 1986b, McCullough et al. 2009). Owing to the increased flight response of *A. auroguttatus* to injured trees in the higher crown classes, girdled trees may be an effective monitoring tool for *A. auroguttatus* populations. The release rate or composition of attractive volatiles from injured trees may decline or change as the trees die and desiccate (see crown class 5, Fig. 4B).

We interpret D-shaped emergence hole densities as an indirect measure of oviposition and subsequent larval development and injury to oak phloem caused by larvae. Similar to *A. auroguttatus* landing rates, emergence hole densities were greater on larger-diameter trees (Fig. 5A) and on trees with higher levels of crown injury (Fig. 5B). There was also a significant trend toward greater annual increment of emergence hole densities with increasing crown rating (Fig. 5D). The measurable emergence hole densities in 2011 and the positive annual increment between 2010 and 2011 in newly dead trees (<1 yr; Figs. 5B and D) likely reflected the delayed development by the last cohort of adults from these trees. The parents of this cohort likely arrived a year or two earlier in large numbers on living, but severely injured, trees that we would have rated in crown class 4 (Fig. 4B). These data highlight the importance of leaving wood from recently killed trees on site for 2 yr as a management practice before transporting it for use as firewood (Jones et al. 2013).

As injury from *A. auroguttatus* larvae increases, the host tissue likely becomes more favorable (e.g., changes in phloem chemistry and water content) for immature populations as has been observed with *A. planipennis* (Chen et al. 2011). Thus, oaks that are currently showing moderate to severe injury by *A. auroguttatus* in southern California are likely to succumb to increased herbivory by this pest in the next few years. Felling and mechanical grinding (<7.6 cm particle size) of these heavily infested trees may be the best option to reduce local populations, especially in areas with low population densities (McCullough et al. 2009, Jones et al. 2013). The increases in both *A. auroguttatus* landing rates and emergence hole densities across the crown classes validate the tree health rating system developed by Coleman et al. (2011), and its use as a decision tool for implementing management options.

Our seasonal flight data from the various experiments reported in this article are consistent with the collection history of *A. auroguttatus* and with other observations by our group. In a survey of museum collection records, adult *A. auroguttatus* were taken by collectors between 12 June and 14 September (based on 28 specimens with dates recorded on the labels, Coleman and Seybold 2011). These records included seven specimens in June, 12 specimens in July, seven specimens in August, and one specimen in September. With closely related *Agrilus coxalis* (also 28 dated collection records, Coleman and Seybold 2011), adults were taken in Mexico and Central America between 21 March and 28 September with one specimen in March (possibly mislabeled), nine specimens in May, seven specimens in June, nine specimens in July, and two specimens in September. This general adult activity recorded by collectors is consistent with our data on adult *A. auroguttatus* flight, which began in mid-May and continued until early to mid-September across our various trapping site locations. In 2009, *A. auroguttatus* adults were not detected on prism flight-intercept traps deployed at higher elevations (1,800 m) until the week of 29 May. Adult flight peaked in mid-June to early July, and this peak flight interval was supported in multiple years by each of the three trapping studies assessing flight periodicity and by previous studies in San Diego Co. (Coleman and Seybold 2008b; Haavik et al. 2013). Adult trap catch diminished (<10 beetles per week) on all trap types by the beginning of August and remained low through September. *A. auroguttatus* flight activity has been recorded into November with purple prism flight-intercept traps, but in that instance, trap catches were extremely low (Coleman and Seybold 2008b). The flight data from the current study are also consistent with the emergence periods of *A. auroguttatus* from infested firewood in the field (Jones et al. 2013). The flight data for *A. auroguttatus* support a univoltine life cycle in southern California. Trapping studies or traps used for monitoring the distribution of *A. auroguttatus* in southern California should begin by mid-May and continue to early September to capture the majority of the flight period.

The delimitation survey effectively detected *A. auroguttatus* populations in known infested areas and populations on the edge of the infestation in San Diego Co. Traps were not effective at detecting a satellite population in 2012 in eastern Riverside Co. (Jones et al. 2013). However, the closest traps (San Bernardino National Forest, San Jacinto Ranger District, and Black Mountain Rd.) were ≈7 km from the infested area in Idyllwild, CA. The results of the delimitation survey support the hypothesis that *A. auroguttatus* populations were introduced into California and that their distribution is limited, but expanding (Coleman et al. 2012a).

Visual and olfactory cues appear to play an important role in determining a suitable host for *A. auroguttatus* adults, similar to *A. planipennis* (Crook and Mastro 2010). Future trapping studies for *A. auroguttatus* should incorporate both cues. Conducting electroretinogram assays and measuring leaf and bark reflectance can assist with refining an effective trap color for *A. auroguttatus* (Crook and Mastro 2010). Identifying semiochemical components for effective lures will also assist in future ecological studies, insecticide trials, integrated pest management efforts, and delimiting the distribution of *A. auroguttatus* in California.

Acknowledgments

We thank Andreana Cipollone, Joel Egan, and Grayland Walter, USDA Forest Service, Forest Health Protection; Zachary Heath, USDA Forest Service, Forest Health Monitoring; Michael I. Jones, University of California, Davis; Kim Camilli, CA Department of Forestry and Fire; and the Cleveland National Forest for their assistance with and support of this work. Jason Oliver, Tennessee State University, McMinnville, and three anonymous reviewers graciously provided comments on previous versions of this manuscript. Funding for this work was provided by the USDA Forest Service, Forest Health Protection, Pacific Southwest Region; by the USDA Forest Service Pacific Southwest Research Station Invasive Species Program; by the USDA Forest Service, Forest Health Monitoring Program, Detection Monitoring (Project #WC-DM-09-01) and Evaluation Monitoring (Project #WC-EM-F-10-01); USDA Forest Service Special Technology Development Program (Project #R5-2011-03); and by the USDA Animal and Plant Health Inspection Service (APHIS) Center for Plant Health Sciences and Technology (Agreements #1081001528CA and 1181301528CA). We are especially grateful to Vic Mastro for providing and facilitating the support from USDA APHIS.

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Received 27 May 2013; accepted 12 March 2014.