Testing Baits to Control Argentine Ants (Hymenoptera: Formicidae) in Vineyards

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ABSTRACT Liquid baits were evaluated for control of the Argentine ant, *Linepithema humile* (Mayr) (Hymenoptera: Formicidae), and associated mealybug and soft scale pests in California vineyards. In 2003, liquid baits with small doses of imidacloprid, boric acid, or thiamethoxam dissolved in 25% sucrose water resulted in lower ant and mealybug densities and fruit damage, compared with an untreated control. Similar treatments in a soft scale-infested vineyard showed only a reduction of ant density and fruit infestation in only the boric acid and thiamethoxam treatments. In 2004, commercial and noncommercial formulations of liquid baits reduced ant densities in three separate trials, but they had inconsistent effects on mealybug densities and fruit infestation; granular protein bait had no effect. Using large plots and commercial application methodologies, liquid bait deployed in June resulted in lower ant density and fruit infestation, but it had no effect on mealybug density. Across all trials, liquid bait treatments resulted in lower ant density (12 of 14 trials) and fruit damage (11 of 14 sites), presenting the first report of liquid baits applied using commercial methodologies that resulted in a reduction of ants and their associated hemipteran crop damage. For commercialization of liquid baits, we showed that any of the tested insecticides can suppress Argentine ants when properly delivered in the crop system. For imidacloprid, bait dispensers must be protected from sunlight to reduce photodegradation. Results suggest that incomplete ant suppression can suppress mealybug densities. However, after ant populations are suppressed, there may be a longer period before hemipteran populations are effectively suppressed. Therefore, liquid baits should be considered part of a multiseason program rather than a direct, in-season control of hemipteran pest populations.

KEY WORDS Argentine ant, *Linepithema humile*, Pseudococcidae, liquid baits, vineyards

The Argentine ant, *Linepithema humile* (Mayr) (Hymenoptera: Formicidae), is an invasive species that has established over a wide geographic range, often with damaging economic and ecological effects (Holway et al. 2002). Although commonly recognized as an urban pest (Rust and Knight 1990), the Argentine ant also has adverse impacts in natural and agricultural systems. In most introduced regions, its unicolonial nest structure, low genetic diversity, high population density, and efficient use of resources can provide a competitive advantage over other ant species (Chen and Nonacs 2000, Tsutsui et al. 2003). As a result, Argentine ants often displace native ants, and their presence can disrupt other invertebrates and even vertebrate and plant populations (Sanders et al. 2001, Suarez et al. 2002). In agricultural systems, Argentine ants are associated with outbreaks of phloem-feeding hemipterans, which they tend and protect from natural enemies in return for honeydew, a nutrient-rich food source (Buckley and Gullan 1991).

In California's coastal wine vineyards, the Argentine ant has been implicated in outbreaks of the grape mealybug, *Pseudococcus maritimus* (Ehrhorn), and the obscure mealybug, *Pseudococcus viburni* (Signoret) (Daane et al. 2007). Mealybugs infest fruit clusters and damage the vine by shunting photosynthates from the phloem and by excreting honeydew, which promotes the growth of sooty molds that further inhibit photosynthesis. Mealybugs also can lower crop value by transmitting closteroviruses (Golino et al. 1999.). Argentine ants also tend soft scales (Buckley and Gullan 1991), and they may be associated with increasing problems with European fruit lecanium, *Parthenolecanium corni* (Bouché), in vineyards.

Exclusion experiments using sticky barriers on the vine trunk have shown that complete removal of Argentine ants can lower both grape and obscure mealybug densities and their crop damage (Daane et al. 2007). However, commercial use of sticky barriers to exclude ants in California vineyards is impractical because of high application costs. In lieu of commercially

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effective cultural or biological controls, research has instead focused on insecticides. Historically, Argentine ant control has relied on contact insecticides that act as barrier treatments (Rust 2001; Klotz et al. 2002, 2003). These sprays provide only partial suppression: although they kill foragers, they have little or no effect on the queens and brood in the nest (Knight and Rust 1990). Foragers constitute only a small fraction of the workforce, and they are quickly replaced by nestmates that mature during the treatment period. Another drawback of the chemical barriers is that they commonly degrade within 30 d, increasing the need for repeated applications (Rust et al. 1996).

Containerized ant baits present several advantages over traditional barrier sprays. First, baits exploit ant foraging and social behaviors as scouts recruit nestmates to bait stations (Silverman and Roulston 2001). Argentine ants use persistent trail pheromones to orient colony members to food resources, resulting in fidelity to bait station locations (Vega and Rust 2001). Because bait is exchanged among colony members (including larvae and queens) via trophallaxis (Knight and Rust 1991), baits have the potential to provide season-long control, rather than a temporary reduction of foragers. Baits also reduce undesirable environmental impacts, because only a small amount of insecticide is placed in the field (Taniguchi et al. 2005). Ant baits are prepared in two main forms: granular (dry) protein or soybean oil baits, and liquid sucrose-based baits. The granular baits have been shown to be effective against fire ants and other myrmicine ants in various systems, including nursery (Costa et al. 2001), forest (Krushelnycky and Reimer 1998), urban (Blachly and Forschler 1996, Klotz et al. 2002), and agriculture (Taniguchi et al. 2005, Tollerup et al. 2005). However, they are not highly attractive to Argentine ants (Baker et al. 1985, Klotz et al. 2002), whose diet consists mainly of sugars. For example, >99% of the material carried to the nest by Argentine ants foraging on citrus trees was either citrus nectar or honeydew produced by the citrus mealybug, *Planococcus citri* (Risso) (Markin 1970b). This preference required the development of liquid, carbohydrate baits for Argentine ant control (Silverman and Roulston 2001, Rust et al. 2004). We previously demonstrated that liquid baits with a low dose of thiamethoxam (0.0001%) can reduce Argentine ant densities in California vineyards and that this reduction results in suppression of mealybugs and their crop damage (Daane et al. 2006). Here, we report on continued research toward the development of an ant control program for vineyards, with the specific goal of testing insecticide materials and using application techniques that might be suitable for commercial operations.

Materials and Methods

Field Sites. Experiments were conducted in California's North Coast and Central Coast wine grape regions. All vineyards had populations of Argentine ants and associated hemipteran pests. The North Coast vineyards were infested with either the grape mealybug or the European fruit lecanium scale. The Central Coast vineyards were infested with the obscure mealybug. All vineyards were mature (>8 yr old), on dripline irrigation systems, and with vine spacings that resulted in 1,282–2,240 vines per ha. Vineyard floors were cover cropped in alternate row middles (Central Coast) or all row middles (North Coast). A herbicide (glyphosate) was applied for weed management within rows. During the experiment, no insecticides were used (other than the applied treatments), but sterol inhibitors and/or sulfur were applied to control powdery mildew (*Uncinula necator* Burrill).

Insect Sampling. Ant densities were determined by a measurement of feeding activity, assessed as the amount of nontoxic sucrose water removed from 50-ml polypropylene centrifuge tubes (Corning Inc., Corning, NY) tied to the vine trunk (Klotz et al. 2002). The 50-ml tubes are henceforth referred to as monitoring tubes. A 2-cm hole was drilled in the cap, and a square of permeable plastic mesh (Weedblock, Easy Gardener Inc., Waco, TX), was placed between the cap and the filled tube, covering the hole. The mesh was fine enough to retain the liquid when the tube was inverted, but it was coarse enough to allow ants to remove the liquid on contact. The monitoring tubes were filled to 45 ml with 25% sucrose water, weighed, and inverted on a vine trunk for 24–72 h (depending on ant activity). They were then brought back to the laboratory and reweighed. One additional monitoring tube per plot was attached to an ant-excluded bamboo stake to measure the amount of water lost to evaporation, which was averaged across all plots and used to adjust the final weight. Because each milliliter of sugar water emptied from the monitoring tube represents ≈3,300 ant visits (Vega and Rust 2001), sugar water removal rates are related to ant density.

Mealybugs and European fruit lecanium scale were monitored using timed visual searches, based on methods developed by Geiger and Daane (2001). Randomly selected vines were searched for 2.5 or 3 min each, and the numbers of second instar to adult stage mealybugs or scales were recorded. An experienced sampler could determine that part of vine where these pests were most likely to occur at a given time of year, which allowed adjustment for seasonal changes in their distribution on the vine. Bark on the trunk, cordon, and spurs was stripped away when necessary. Because the visual search was a destructive process, individual vines were sampled only once during the season.

To assess crop damage at harvest time, vines were randomly selected within each plot, and three fruit clusters from each of the selected vines were rated at harvest on a 0–3 scale, where 0 represents no mealybugs; 1 indicates presence of honeydew and/or ≤10 mealybugs; 2 represents >10 mealybugs, sooty mold, and/or honeydew; and 3 indicates heavily infested, unmarketable clusters. Fruit clusters in direct contact with woody parts of the vine were preferentially sampled, when available, because such clusters are more likely to be infested with mealybugs (Geiger and Daane 2001). Fruit damage levels address economic damage, and they may also be a better assessment of mealybug density than the timed mealybug counts because more vines could be sampled during the same period. For each sampling method used, the sampling frequency and number of vines sampled are provided for each experimental trial.

Bait Dispensers. The bait dispensers were made from 250-ml polypropylene centrifuge tubes (Corning Inc.). They were filled to 240 ml with bait solution, and they were deployed on the vine as described above for the monitoring tubes. The tubes were cleaned and refilled every 2–3 wk throughout each experiment. In 2004 experiments, modifications to the bait dispenser were made for two insecticide materials. First, we suspected that imidacloprid was susceptible to photodegradation in the bait dispensers; the 2004 treatments with imidacloprid were shielded with Styrofoam containers (5 by 7 by 17 cm) (Amerifoods Trading Co., Los Angeles, CA). The bait dispensers fit inside the containers, whereas the cap remained exposed to allow ants to feed. Second, when testing granular spinosad, pellets were loaded into bait dispensers that were then deployed horizontally on the vine and without the plastic mesh, allowing ants' access to the granular bait.

Experiment 1: Active Ingredients. In 2003, the effectiveness of three active ingredients and an untreated control were tested. Active ingredients were 0.5% crystalline boric acid (Fisher Chemicals, Fair Lawn, NJ), 0.0001% imidacloprid (Bayer CropScience, Kansas City, MO), and 0.0001% thiamethoxam (Syngenta Crop Protection, Richmond, CA). All of the percentages are weight to volume in water, and each chemical was technical grade material. The bait/food was 25% sucrose water. Boric acid was mixed directly into sucrose water; thiamethoxam and imidacloprid were first dissolved in 1–2 ml of ethanol, and then they were mixed into the sucrose water (15–20 liters). Dilute quantities of ethanol have no effect on the consumption of sucrose water by Argentine ants (Baker et al. 1985).

Experiments were conducted in two independent sites: a Napa Valley appellation Chardonnay vineyard (8.1 ha) (38° 12′20.68″ N, 122° 14′08.17″ W) infested with the European fruit lecanium scale and an Edna Valley appellation Chardonnay vineyard (10 ha) (35° 13′48.13″ N, 120° 35′34.83″ W) infested with the obscure mealybug. The four treatments were established in a randomized complete block design, with four replicates per treatment at each site. Each replicate was a 200 vine plot (five rows × 40 vines); row/vine spacing was 2.44 × 1.83 m (2240 vines per ha) and 3.05 × 1.83 m (1793 vines per ha), respectively, for the Napa and Edna Valley plots. Neighboring replicates were separated by five row buffers (no plots were within the same rows). The bait dispensers were deployed on 7 and 14 April at Edna Valley and Napa Valley appellation vineyards, respectively, at a density equivalent to 123 dispensers per ha.

Ant density was measured at 2-wk intervals, from January through October, by using 17 monitoring tubes and one evaporation tube per replicate. Scale or mealybug counts were made approximately every 30 d, from February to October, on 10 randomly selected vines per replicate, selected from the middle rows of each plot. Crop damage was evaluated at the Napa Valley site on 16 September, by using 20 vines per replicate (10% of the vines), and at the Edna Valley site on 14 October, by using 12 vines per replicate (6% of the vines).

Experiment 2: Commercial and Novel Formulations. In 2004, treatments were 1) a commercial ant bait of 0.005% imidacloprid, delivered in a liquid sucrose solution (Pre-Empt, Bayer CropScience); 2) a commercial ant bait of 0.015% spinosad, delivered in a granular bait (Justice, Dow AgroSciences, Indianapolis, IN); 3) a noncommercial formulation of 0.015% spinosad (technical grade, Dow AgroSciences), delivered in liquid sucrose bait; and 4) an untreated control. Pre-Empt was tested using an experimental use permit as it was registered for urban but not agricultural use; Bayer CropScience is currently marketing Vitis (0.001% imidacloprid) for commercial use in vineyards. Three independent experiment sites were established: a Sonoma Pinot Noir vineyard (14.2 ha) (38° 15′11.27″ N, 122° 26′50.11″ W), a Santa Maria Gewürztraminer vineyard (45.8 ha) (34° 48′41.04″ N, 120° 26′39.96″ W), and a Santa Maria appellation Chardonnay vineyard (29.7 ha) (34° 52′57.98″ N, 120° 15′51.64″ W). At each site, the four treatments were set in a randomized complete block design with three replicates. Each replicate was an 80 vine plot (four rows × 20 vines); row/vine spacing was 3.05 × 1.83 m (1793 vines per ha) at the Sonoma and Santa Maria Gewürztraminer sites, and 3.66 × 2.14 m (1,282 vines per ha) at the Santa Maria Chardonnay site. There were five-row buffers between each replicate. Bait stations were deployed at a rate of 10 per plot, which was equivalent to 160 and 223 per ha (1,282 and 1,793 vines per ha, respectively), on 14 April (Pinot Noir), 31 March (Gewürztraminer), and 2 April (Chardonnay).

Ant density was monitored approximately every 30 d from April through October 2004, by using 10 monitoring tubes and one evaporation tube per replicate. Timed mealybug counts were made approximately every 30 d on five vines per replicate. Crop damage was evaluated on eight vines per replicate (10% of the vines) on 16 August (Pinot Noir) and 1 and 13 September (Gewürztraminer and Chardonnay, respectively).

Experiment 3: Imidacloprid in Large Plots. In 2003, an experiment was conducted to determine the effect of placing fewer ant baits over a larger area. This experiment addressed our concerns that there was too much ant movement between treatments in experiments using smaller plots (e.g., four rows × 20 vines, five-row buffers) and that the bait station deployment rate (e.g., up to 223 per ha) was not commercially feasible. This experiment used a randomized complete block design with two plots in each of five blocks. Each block was located in a different vineyard, each infested with Argentine ants and obscure mealybugs. The minimum distance between vineyards was 2 km.

The treatments (bait and control) were assigned to the two plots in each block; the minimum distance separating the plots was 50 m.

Bait stations were filled with 0.0001% imidacloprid in 25% sucrose water and deployed at a density of 123 per ha, spaced evenly throughout each plot. Bait plots were relatively large (12 rows × 35 vines to 24 rows × 60 vines, or ≈0.5–1.1 ha each), whereas the control plots were smaller (five rows × 35 vines, 0.15 ha) to accommodate the collaborating vineyard managers' concern that a limited area would be placed at risk without any ant or mealybug controls. Areas outside of the bait and control plots were treated as needed with insecticides. The appellation, cultivar, and bait station deployment periods in the five vineyards were as follows: a Santa Ynez appellation Chardonnay block (34° 43′08.24″ N, 120° 08′29.31″ W) with baits deployed on 8 May, a Santa Maria Gewürztraminer block (34° 48′41.04″ N, 120° 26′39.96″ W) with baits deployed on 14 May, two Santa Maria appellation Chardonnay blocks (34° 52′57.98″ N, 120° 15′51.64″ W) with baits deployed on 16 and 17 June, and an Edna Valley appellation Chardonnay block (35° 13′23.40″ N, 120° 35′36.87″ W) with baits deployed on 3 July.

Ant density was monitored three times during the growing season (July, August, and September), by using 18 monitoring tubes and one evaporation tube per replicate. Timed mealybug counts were taken three times (preharvest counts, 1–12 July and 12–18 August and postharvest counts, 15 September–6 October) on 30 and 15 randomly selected vines per plot in the bait and control treatments, respectively. The difference in sample dates reflected the logistics of sampling geographically separated plots and differences in vine phenology; the difference in the number of sampled vines reflected differences in plot sizes. Crop damage was evaluated between 4 September and 15 October 2003, on 10% of the vines per treatment plot.

Experiment 4: Thiamethoxam Bait and Sticky Barriers. In 2003, an experiment was conducted to compare the effects of ant suppression using liquid bait versus complete ant exclusion. The vineyard was a 10.7-ha Pinot Noir block in the Carneros appellation (38° 14′09.49″ N, 122° 23′10.84″ W) that was infested with the grape mealybug; the experimental area was 50 vines × 85 rows divided into eight plots. Plots were 50 vines × 5 rows, with five row buffers between plots; row/vine spacing was 2.44 m × 1.83 m (2240 vines per ha). Treatments were liquid bait (0.0001% thiamethoxam) in 25% sucrose water and ununtreated control. Bait and control treatments were set in a randomized complete block design, with four replicates. Within each of the bait treatment plots, five ant-excluded vines were established (20 total). To exclude ants, shoots were pruned back from adjacent vines to isolate the vine and the trellis-wires, and the base of the trunks was swabbed with Stikem resin (Seabright Laboratories, Emeryville, CA). The sticky barriers were checked every 2 wk from May through September, and the Stikem barriers were reapplied as needed. Bait dispensers were dispersed at 10 vine intervals, which was equivalent to 123 tubes per ha, and they were deployed 7 January.

Ant feeding activity was monitored every 2 wk using 23 monitoring tubes and one evaporation tube per replicate. Ant monitoring tubes were not placed on the exclusion vines, but instead a visual count of all ants moving past a fixed point on the vine trunk was made during a 30-s period every 2–4 wk, from March to October. Timed mealybug counts were made every 4 wk, from February to October, on 10 randomly selected vines per replicate. The exclusion vines could not be repeatedly sampled using the timed counts because of the destructive nature of the procedure; thus, they were sampled only once, after harvest. Crop damage was evaluated on 5 September on 30 vines per plot (12% of the vines), and on all the exclusion vines.

Statistics. Results are presented as means per treatment (±SEM). Season-long treatment effects on insect densities were determined using a general linear model. The model used ant feeding activity (grams of sucrose water removed per day) or timed visual counts (insects per vine per timed count) as a function of treatment, sample date, and treatment × sample date interaction. Sample date was set as a categorical variable to exclude its effect on treatment. If the treatment × sample date interaction term is not significant ($P > 0.05$), this analysis is equivalent to an analysis of covariance with sample date as the covariate. For all analyses, the average per replicate was used as the sample unit. Data were transformed ($\log[x + 1]$) as needed to stabilize the variance. Results of the general linear model are summarized in *Appendix* 1 and 2. When there was a treatment effect, treatments were separated using Scheffé's test; the pairwise comparisons are discussed in the text. For fruit damage ratings the treatment effects were compared using contingency tables (Systat Software, Inc. 2007). For all experiments with three or more treatments, pairwise comparisons (treatment × damage ratings) were made for all possible treatment combinations, with an experiment-wide error rate at $\alpha = 0.05/n$, where n is the number of possible pairwise comparisons.

Results

Experiment 1: Active Ingredients. Season-long ant density differed among treatments at the Napa Valley site (Fig. 1A); pairwise comparisons showed less sucrose removed (lower ant density) from monitoring tubes in the boric acid than control and imidacloprid treatments ($P < 0.001$ and $P = 0.002$, respectively). Ant density in the thiamethoxam treatment was also lower than in the control and imidacloprid treatments ($P < 0.001$ and $P = 0.009$, respectively). There was no difference between boric acid and thiamethoxam ($P = 0.981$) or between the control and imidacloprid ($P = 0.159$) treatments.

There was no treatment effect on season-long European fruit lecanium densities ($F = 1.920$, df = 3, $P = 0.135$). Fruit damage, however, corresponded closer to ant density, with lower ratings in the boric acid and thiamethoxam treatments compared with the control,

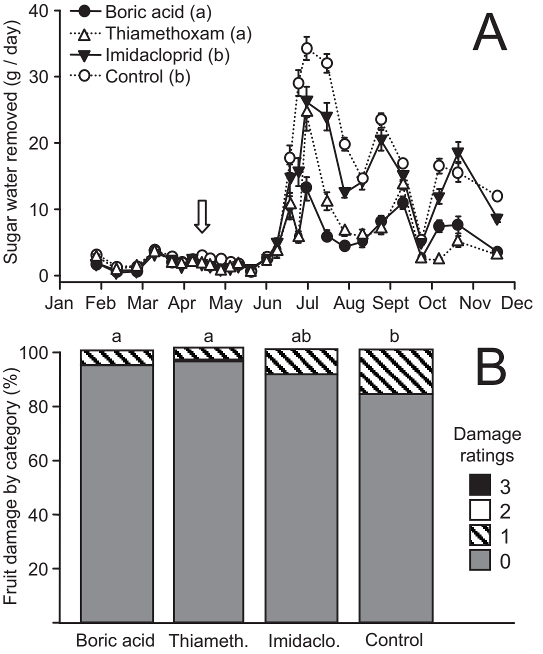

Fig. 1. Ant feeding activity (A) and fruit damage (B) in a Napa Valley appellation vineyard, infested with European fruit lecanium scale, from a 2003 experiment comparing liquid baits with different active ingredients. Arrow indicates when baits were first deployed. Table 1 provides the general linear model for ant feeding activity and pairwise comparisons are provided in the text. Fruit damage ratings differed among treatments ($\chi^2 = 12.12$, df = 3, $P = 0.007$); pairwise comparisons (df = 3, $\alpha = 0.01$) are as follows: control versus boric acid: $\chi^2 = 6.750, P = 0.0094$; control versus imidacloprid: $\chi^2 = 3.485$, $P = 0.0654$; control versus thiamethoxam: $\chi^2 = 8.223$, $P = 0.0041$; boric acid versus imidacloprid: $\chi^2 = 0.645$, $P = 0.4220$; boric acid versus thiamethoxam: $\chi^2 = 0.096$, $P = 0.7569$; and imidacloprid versus thiamethoxam: $\chi^2 = 1.222$, $P = 0.2689$. Different letters in parentheses (insect densities) or above each bar (fruit damage) indicate significant differences between treatments.

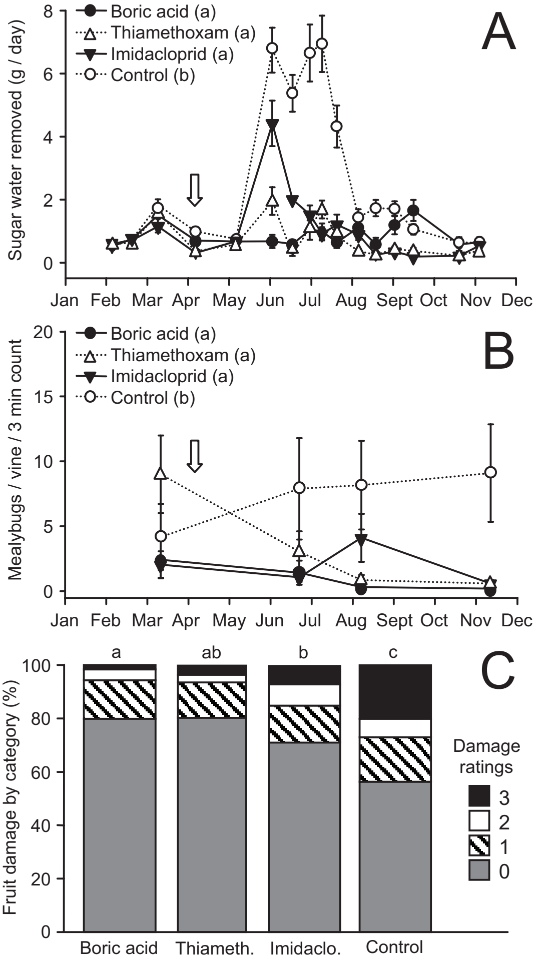

Fig. 2. Ant density (A), mealybug density (B), and fruit damage (C) in an Edna Valley appellation vineyard, infested with obscure mealybug, from a 2003 experiment comparing liquid baits with different active ingredients. Arrow indicates when baits were first deployed. For ant and mealybug densities, the general linear model output is provided in *Appendix* 2, and pairwise comparisons are provided in the text. Fruit damage ratings differed among treatments ($\chi^2 = 99.57$, df = 9, $P < 0.001$); pairwise comparisons (df = 3, $\alpha = 0.01$) are as follows: boric acid versus thiamethoxam, imidacloprid, control: $\chi^2 = 1.613$, $P = 0.653$; $\chi^2 = 13.72$, $P = 0.003$; $\chi^2 = 60.34$, $P < 0.001$; thiamethoxam versus imidacloprid and control: $\chi^2 = 10.58$, $P = 0.014$ and $\chi^2 = 48.64$, $P < 0.001$, respectively; and imidacloprid versus control: $\chi^2 = 27.22$, $P < 0.001$. Different letters in parentheses (insect densities) or above each bar (fruit damage) indicate significant differences between treatments.

whereas the imidacloprid was not different from other treatments (Fig. 1B).

At the Edna Valley site, ant densities were lower in the boric acid, thiamethoxam, and imidacloprid treatments compared with the control ($P < 0.001$ for each comparison). Ant density in the boric acid treatment was not different from either thiamethoxam or imidacloprid ($P = 0.986$ and $P = 0.999$, respectively), nor was there a difference between thiamethoxam and imidacloprid ($P = 0.964$) (Fig. 2A). Similar to ant densities, mealybug densities in the boric acid, thiamethoxam, and imidacloprid treatments were significantly lower than in the control ($P < 0.001$, $P = 0.009$, $P = 0.005$, respectively) (Fig. 2B). There were no other treatment effects on mealybug densities (pairwise comparisons of boric acid versus thiamethoxam and imidacloprid were $P = 0.751$ and $P = 0.\ 847$, respectively, and thiamethoxam versus imidacloprid was $P = 0.998$). Fruit damage ratings were lower in all bait treatments than in the control, and were lower in the boric acid than in the imidacloprid treatment (Fig. 2C).

Experiment 2: Commercial Formulations. At the Sonoma Valley vineyard, ant densities in plots with liquid formulations of imidacloprid and spinosad were lower than those in the granular spinosad and control treatments ($P < 0.001$, for each comparison), whereas there was no difference between the liquid imidaclo-

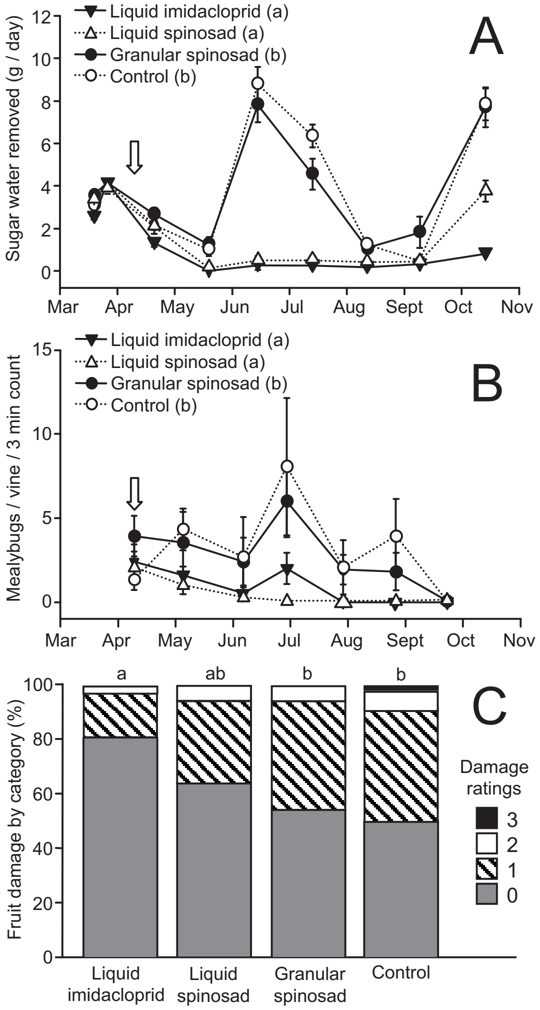

Fig. 3. Ant density (A), mealybug density (B), and fruit damage (C) in a Sonoma appellation vineyard, infested with grape mealybug, from a 2004 experiment comparing liquid formulations of imidacloprid and spinosad, a granular formulation of spinosad, and an untreated control. Arrow indicates when baits were first deployed. For ant and mealybug densities, the general linear model output is provided in *Appendix* 2, and pairwise comparisons are provided in the text. Fruit damage ratings differed among treatments ($\chi^2 = 19.57$, df = 9, $P = 0.021$); pairwise comparisons (df = 3, $\alpha = 0.01$) are as follows: liquid imidacloprid versus liquid spinosad, granular spinosad, and control: $\chi^2 = 4.992$, $P = 0.082$; $\chi^2 = 11.44$, $P = 0.003$; $\chi^2 = 15.14$, $P = 0.002$, respectively; liquid spinosad versus granular spinosad and control: $\chi^2 = 1.537$, $P = 0.464$ and $\chi^2 = 3.561$, $P = 0.313$, respectively; and granular spinosad versus control: $\chi^2 = 1.248$, $P = 0.741$. Different letters in parentheses (insect densities) or above each bar (fruit damage) indicate significant differences between treatments.

prid and spinosad ($P = 0.375$) or granular spinosad and control ($P = 0.998$) treatments (Fig. 3A). Mealybug density showed a similar pattern, with lower counts in liquid formulations of imidacloprid and spinosad than the control ($P = 0.037$ and $P = 0.002$, respectively) and the granular spinosad ($P = 0.048$ and $P = 0.013$, respectively) treatments, and no difference between the liquid formulations of imidacloprid and spinosad ($P = 0.802$) or the granular spinosad and control ($P = 0.997$) treatments (Fig. 3B). Treatment effect on fruit damage followed a slightly different pattern, with lower damage in the liquid imidacloprid than in both the control and granular spinosad treatments, but no difference between liquid spinosad and the other treatments (Fig. 3C).

At the Santa Maria Gewürztraminer vineyard, ant densities were lower in liquid formulations of imidacloprid and spinosad than in the granular spinosad and control treatments ($P < 0.001$ for all pairwise comparisons), whereas there was no difference between the two liquid formulations ($P = 0.728$) or between the granular spinosad and control ($P = 0.969$) treatments (Fig. 4A). Mealybug densities followed a different pattern, with lower counts only in treatments with liquid formulations of imidacloprid and spinosad compared with the granular spinosad ($P = 0.008$ and $P = 0.011$, respectively), whereas all other pairwise comparisons showed no treatment effect (control versus liquid imidacloprid, liquid spinosad, and granular spinosad were $P = 0.679$, $P = 0.751$, and $P = 0.111$, respectively; liquid formulations of imidacloprid versus spinosad was $P = 0.999$) (Fig. 4B). Fruit damage was lower in the liquid imidacloprid than the granular spinosad and control treatments; there were no other statistically significant pairwise comparisons (Fig. 4C).

At the Santa Maria Chardonnay vineyard, there were few treatment effects. Ant density was lower only in the liquid imidacloprid treatment compared with the control ($P = 0.\ 022$) (Fig. 5A), with no other significant pairwise comparisons (liquid imidacloprid versus liquid spinosad and granular spinosad were $P = 0.151$ and $P = 0.312$, respectively; liquid spinosad versus granular spinosad and control were $P = 0.970$ and $P = 0.861$, respectively; and granular spinosad versus control was $P = 0.607$). Similar to ant density patterns, mealybug density was lower only in the liquid imidacloprid treatment compared with the control ($P = 0.\ 020$) (Fig. 5B), with no other differences between other pairwise comparisons (liquid imidacloprid versus liquid spinosad and granular spinosad were $P = 0.381$ and $P = 0.115$, respectively; liquid spinosad versus granular spinosad and control was $P = 0.924$ and $P = 0.582$; and granular spinosad versus control was $P = 0.918$). In contrast, fruit damage ratings were lower in all bait treatments than in control, and fruit damage was also lower in the granular spinosad than other bait treatments (Fig. 5C).

Experiment 3: Imidacloprid in Large Plots. Ant density was lower in the imidacloprid bait treatment than in the control ($F = 24.94$; df = 1,2,2; $P < 0.0001$; Fig. 6A). There was no treatment effect on mealybug densities ($F = 0.317$; df = 1, 2, 2; $P = 0.578$); however, fruit damage was lower in the bait than control treatment ($\chi^2 = 11.61$, $P < 0.001$) (Fig. 6B).

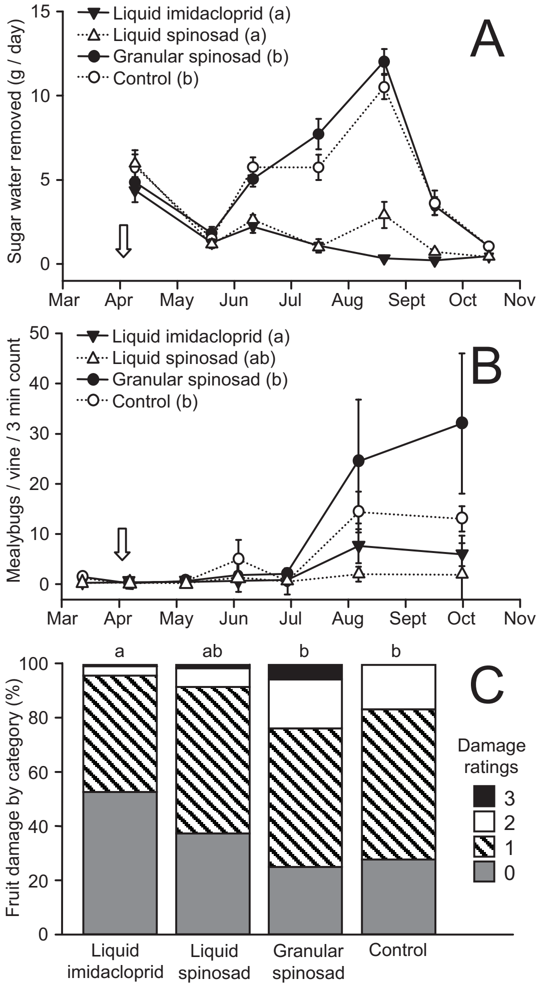

Fig. 4. Ant density (A), mealybug density (B), and fruit damage (C) in a Santa Maria appellation Gewürztraminer vineyard, infested with obscure mealybug, from a 2004 experiment comparing liquid formulations of imidacloprid and spinosad, a granular formulation of spinosad, and an untreated control. Arrow indicates when baits were first deployed. For ant and mealybug densities, the general linear model output is provided in *Appendix* 2, and pairwise comparisons are provided in the text. Fruit damage ratings differed among treatments ($\chi^2 = 27.58$, df = 9, $P = 0.001$); pairwise comparisons (df = 3, $\alpha = 0.01$) are as follows: liquid imidacloprid versus liquid spinosad, granular spinosad, and control: $\chi^2 = 4.06$, $P = 0.254$; $\chi^2 = 17.5$, $P < 0.001$; $\chi^2 = 14.87$, $P = 0.002$, respectively; liquid spinosad versus granular spinosad and control: $\chi^2 = 7.21$, $P = 0.065$ and $\chi^2 = 5.79$, $P = 0.447$, respectively; and granular spinosad versus control: $\chi^2 = 4.26$, $P = 0.234$. Different letters in parentheses (insect densities) or above each bar (fruit damage) indicate significant differences between treatments.

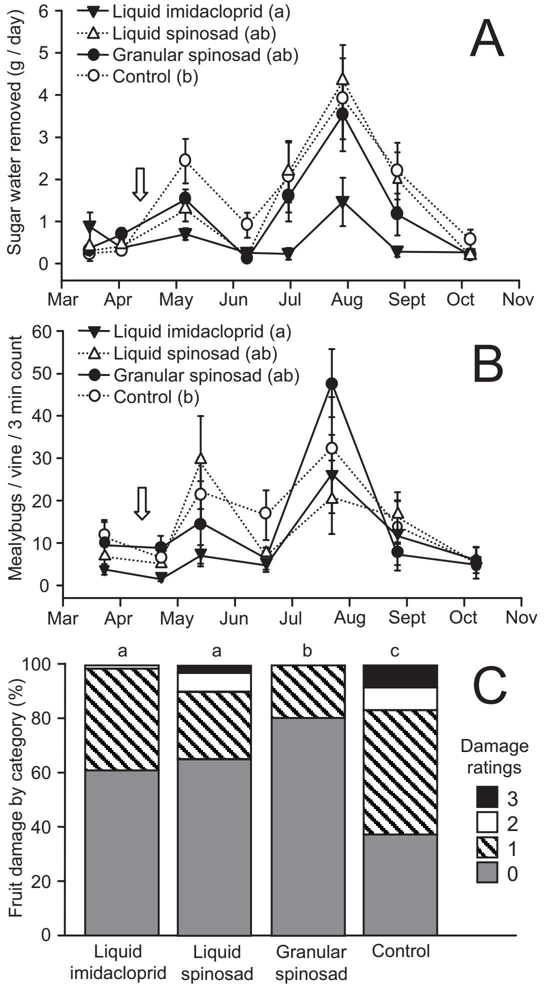

Fig. 5. Ant density (A), mealybug density (B), and fruit damage (C) in a Santa Maria appellation Chardonnay vineyard, infested with obscure mealybug, from a 2004 experiment comparing liquid formulations of imidacloprid and spinosad, a granular formulation of spinosad, and an untreated control. Arrow indicates when baits were first deployed. For ant and mealybug densities, the general linear model output is provided in *Appendix* 2, and pairwise comparisons are provided in the text. Fruit damage ratings differed among treatments ($\chi^2 = 41.54$, df = 9, $P < 0.001$); pairwise comparisons (df = 3, $\alpha = 0.01$) are as follows: liquid imidacloprid versus liquid spinosad, granular spinosad, and control: $\chi^2 = 656$, $P = 0.087$; $\chi^2 = 7.04$, $P = 0.029$; $\chi^2 = 14.24$, $P = 0.002$, respectively; liquid spinosad versus granular spinosad and control: $\chi^2 = 8.65$, $P = 0.034$ and $\chi^2 = 11.9$, $P = 0.008$, respectively; and granular spinosad versus control: $\chi^2 = 31.0$, $P < 0.001$. Different letters in parentheses (insect densities) or above each bar (fruit damage) indicate significant differences between treatments.

Experiment 4: Thiamethoxam and Sticky Barriers. Ant density was lower in thiamethoxam bait than control treatment ($F = 160.7$; df = 1, 28, 28; $P < 0.001$) (Fig. 7A); no ants were observed on the exclusion vines. Mealybug density was lower in the bait than control treatment ($F = 10.27$; df = 1, 8, 8; $P = 0.003$) (Fig. 7B). Mealybug density using the destructive sampling method could not be conducted on the exclusion vines; a postharvest sample of mealybugs showed low mealybug densities on all treatments (<0.3 mealybugs per vine), resulting in no treatment effect ($F = 0.835$; df = 2, 9; $P = 0.465$). However, fruit

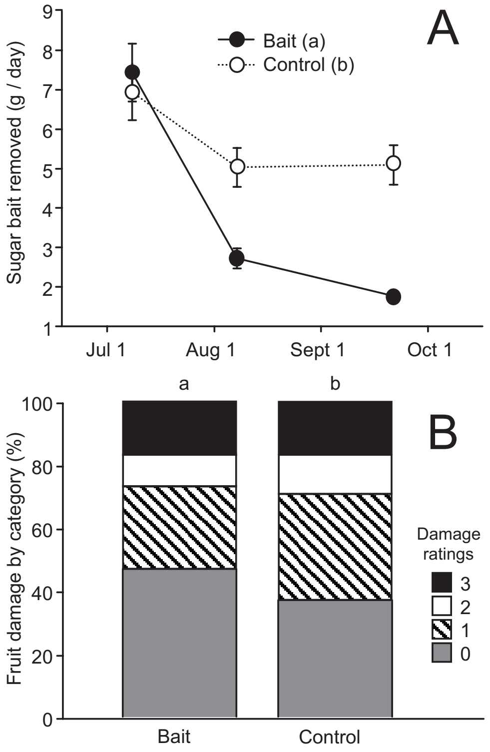

Fig. 6. Mealybug density (A) and fruit damage (B) in treatments from a 2003 experiment in Edna Valley, Santa Maria, and Santa Ynez appellation vineyards comparing a liquid bait containing 0.0001% imidacloprid (technical grade) to an untreated control. Different letters in parentheses (insect densities) or above each bar (fruit damage) indicate significant differences between treatments.

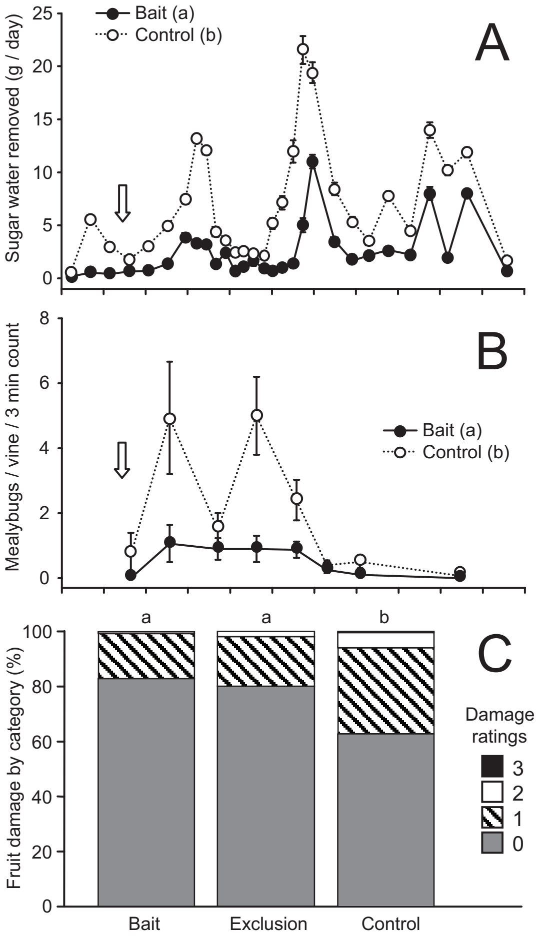

Fig. 7. Ant density (A), mealybug density (B), and fruit damage (C) in a Carneros vineyard, infested with grape mealybug, in treatments comparing liquid bait containing 0.0001% thiamethoxam (technical grade), an untreated control, and vines with a sticky barrier to fully exclude ants. Fruit damage differed among treatments (χ^2 = 44.9, df = 6, $P <$ 0.001); pairwise comparisons (df = 2, α = 0.025) are as follows: follows: control versus bait: χ^2 = 41.86, $P <$ 0.001; control versus exclusion: χ^2 = 10.76, P = 0.013; bait versus exclusion: χ^2 = 2.033, P = 0.362. Different letters in parentheses (insect densities) or above each bar (fruit damage) indicate significant differences between treatments.

damage was lower in the bait and exclusion treatments than the control (Fig. 7C).

Discussion

Experiments reported herein compared the efficacy of insecticide materials that are likely to be registered for use in vineyards and that were deployed using potential commercial methodologies. Three measures of treatment performance were taken: ant density as measured by feeding activity, mealybug or scale density as measured by timed visual counts, and fruit damage as measured by infestation rates at harvest time. Table 1 summarizes treatment effects. Measurements of ant density and fruit damage showed the greatest response to treatments, in part because more of these samples could be collected over a wider area of each plot, thereby reducing the impact of the clumped mealybug distribution (Geiger and Daane 2001). Ant density and fruit damage were lower in treatments with liquid bait, compared with the control in 12 and 11 of 14 trials, respectively (Table 1). Mealybug and scale densities were less responsive to ant bait treatments, with lower hemipteran pest densities in only seven of 14 liquid bait trials. There was no treatment effect on ant or mealybug densities or fruit damage when granular protein bait was tested, as has been shown in other studies (Silverman and Roulston 2001, Rust et al. 2004, but see Krushelnycky and Reimer 1998).

Control options for Argentine ants in agricultural systems are limited by the low number of registered insecticides and lack of biological control agents. This is the first reported successful use of liquid baits in vineyards using commercially viable deployment methodologies. Experimental baits containing fipronil or thiamethoxam provided Argentine ant control in citrus orchards (Klotz et al. 2003) and vineyards (Daane et al. 2006), whereas studies with boric acid

Table 1. Summary of ant bait treatment effects in the four experiments; treatments were significantly lower (L), higher (H) or not different (—) from the control

Trial	Treatment	Site[a]	Pest[b]	Ant density	Pest density	Fruit damage
Exp. 1	Imidacloprid–liquid	Napa	EFLS	—	—	—
		Edna Valley	OMB	L	L	L
	Boric acid–liquid	Napa	EFLS	L	—	L
		Edna Valley	OMB	L	L	L
	Thiamethoxam–liquid	Napa	EFLS	L	—	L
		Edna Valley	OMB	L	L	L
Exp. 2	Imidacloprid–liquid	Sonoma	GMB	L	L	L
		Santa Maria-G	OMB	L	—	L
		Santa Maria-C	OMB	L	L	L
	Spinosad–liquid	Sonoma	GMB	L	L	—
		Santa Maria-G	OMB	L	—	—
		Santa Maria-C	OMB	—	—	L
	Spinosad–granular	Sonoma	GMB	—	—	—
		Santa Maria-G	OMB	—	—	—
		Santa Maria-C	OMB	—	—	—
Exp. 3	Imidacloprid–liquid	Central Coast	OMB	L	—	L
Exp. 4	Imidacloprid–liquid	Carneros	GMB	L	L	L
	Tanglefoot exclusion	Carneros	GMB	L	—	L

[a] Napa Valley Chardonnay (Napa); Edna Valley Chardonnay (Edna Valley); Sonoma Valley Pinot Noir (Sonoma); Santa Maria Gewürztraminer (Santa Maria-G); Santa Maria Chardonnay (Santa Maria-C); Central Coast (Santa Ynez Chardonnay, Santa Maria Gewürztraminer and Chardonnay, and Edna Valley Chardonnay); and Carneros (Carneros Pinot Noir.

[b] Hemipteran pests are European fruit lecanium scale (EFLS), obscure mealybug (OMB), and grape mealybug (GMB).

baits have been limited to organic citrus grooves (Greenberg et al. 2006). Results indicate that any of the tested materials (boric acid, imidacloprid, thiamethoxam, or spinosad) can suppress Argentine ants when properly delivered in liquid sucrose bait. Of the materials tested, imidacloprid, spinosad, and boric acid may be readily registered for use in agricultural systems. Imidacloprid is already registered for use as both foliar and systemic formulations for mealybug and leafhopper (but not ant) in vineyards, and it is one of the more commonly used insecticides in agricultural systems. Spinosad may be of particular interest to some growers because it is an organically registered chemical that is labeled for use on grapes (applied as a foliar) and many other crops. Boron (as the fertilizer boric acid or borate) is a common amendment in vineyards.

Previous studies have shown complete ant exclusion using either insecticides or sticky barriers (e.g., González-Hernández et al. 1999) or experimental liquid bait deployment (Daane et al. 2006) can result in a reduction of hemipteran pests. This is the first report of liquid bait, applied using commercial methodologies, lowering both Argentine ant densities and the associated hemipteran pest crop damage (11 of 14 trials). It is noteworthy that season-long pest densities were lowered in only half of the liquid-bait trials (seven of 14), providing additional information for development of a commercial program. First, the hemipteran pests in the study had relatively long generational times with only one to two (European fruit lecanium scale and grape mealybug) or three to four (obscure mealybug) annual generations. This provided little time for changes in ant densities to effect changes in hemipteran pest densities. Second, their reduction after tending ants are removed is typically associated with higher densities or effectiveness of natural enemies (Daane et al. 2007). At each site, the levels of natural enemies present will vary thus affecting the time needed to impact pest densities. Third, ant exclusion using liquid baits was not complete, immediate, or season-long, (ant reduction was typically greatest in June, July and August). However, we provide evidence that complete, season-long ant exclusion may not be needed to suppress hemipteran pest densities (Fig. 7).

Development of a viable commercial program is also aided by information garnered in those liquid bait trials where there was no effect on ant densities. For example, we found that when the polyurethane bait dispensers were exposed to sunlight there could be photo-degradation of the imidacloprid in solution in only a few hours, which may have contributed to the ineffectiveness of liquid bait in one trial (Fig. 1A). In contrast, there was a reduction in season-long ant densities in other 2003 imidacloprid treatments conducted in the Central Coast region (Figs. 2A and 6A), and we suspect the lower summer temperatures, more foggy days, and a denser vine canopy better protected the bait dispensers from sunlight and reduced the photo-degradation of the insecticide. In the 2004 experiments, in which imidacloprid produced more consistent effects, the bait dispensers were protected from sunlight with Styrofoam containers, which both shielded the solution from light and moderated the temperature of the bait solution, minimizing evaporation and decreasing the frequency at which the baits need refilling.

A concurrent research study showed a significant negative correlation between the density of Argentine ants and bait stations deployed (Nelson and Daane 2007). This is another concern for the commercialization of liquid baits in perennial crop systems: how many stations should be deployed per ha? The large-plot experiment, conducted in five separate vineyards, was designed to test liquid baits dispersed over a larger area and using fewer bait stations per ha. Although there was a reduction in ant activity (Fig. 6A), there was no effect on mealybug density. Importantly, there was considerable fruit damage, with >15% lost crop (Fig. 6B). Although fruit damage in the liquid bait treatment was statistically lower than in the control, this level of crop loss would be unacceptable for most vineyard managers. As mentioned, hemipteran pest densities may not quickly respond to changes in ant densities. In this experiment, a potential flaw in the experimental design was that bait dispensers were deployed relatively late in the season (8 May–3 July). Although liquid baits seemed to have their greatest impact on the summer ant densities, the deployment was possibly too late for effective pest suppression. Brood production for Argentine ants reaches a seasonal high in March (Markin 1970a), with most of this brood directed toward queen development. Similarly, immature mealybugs for the second generation of both grape mealybug (Geiger and Daane 2001) and obscure mealybug (Daane et al. 2007) have been produced by this time, and they can infest fruit clusters as early as July, thereby providing little time for a reduction in ant densities to result in reduced mealybug densities or damage.

Argentine ants have become a key pest in many agricultural systems. Vineyard managers currently have few options other than barrier sprays for their control. The work reported herein summarizes several large field experiments, deploying >20,500 monitoring tubes to measure ant activity and rating >4,600 fruit clusters for pest damage. In aggregate, results showed that small amounts of insecticide delivered in liquid, sucrose baits can suppress Argentine ants in vineyards, which seems to also reduce associated hemipteran pests and their damage to the crop. When properly delivered, all insecticide materials tested were effective. What remains needed for commercialization is information on bait station deployment rate and seasonal periods of deployment, and inexpensive bait stations and bait–insecticide formulations.

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Appendix 1. Relationship of 2003 treatments of different ant bait active ingredients (0.5% boric acid, 0.0001% imidacloprid, 0.0001% thiamethoxam, and an untreated control) and sample date on the season long ant and mealybug or scale densities

Sample	Comparison	Napa Valley[a,b]			Edna Valley[a,b]		
		F-ratio	df	*P* value	*F*-ratio	df	*P* value
Ant density	Treatment	17.039	3	<0.001	14.302	3	<0.001
	Sample date	13.555	26	<0.001	3.772	15	<0.001
	Treatment × date	1.208	78	0.132	1.554	45	0.022
Mealybug or scale density	Treatment	1.920	3	0.135	8.529	3	<0.001
	Sample date	0.981	6	0.445	4.026	3	0.012
	Treatment × date	0.376	18	0.988	1.991	9	0.061

[a] See Figs. 1 and 2 for presentation of data from the Napa Valley and Edna Valley vineyards, respectively.

[b] The Napa Valley vineyard was infested with European fruit lecanium scale, and the Edna Valley vineyard was infested with obscure mealybug.

Appendix 2. Relationship of 2004 treatments of different commercial ant baits (0.15% spinosad [liquid and granular], 0.005% imidacloprid, and an untreated control) and sample date on the season long ant and mealybug densities

Sample	Comparison	Sonoma Valley[a,b]			Santa Maria (Gewürztraminer)[a,b]			Santa Maria (Chardonnay)[a,b]		
		F-ratio	df	*P* value	*F*-ratio	df	*P* value	*F*-ratio	df	*P* value
Ant density	Treatment	41.95	3	<0.001	24.34	3	<0.001	3.666	3	0.016
	Sample date	25.04	8	<0.001	16.12	6	<0.001	10.25	7	<0.001
	Treatment × date	6.656	24	<0.001	4.263	18	<0.001	0.811	21	0.697
Mealybug density	Treatment	8.069	3	<0.001	7.239	3	<0.001	3.653	3	0.013
	Sample date	4.924	6	<0.001	59.15	6	<0.001	5.705	6	<0.001
	Treatment × date	0.941	18	0.536	1.460	18	0.141	0.695	18	0.817

[a] See Figs. 3, 4, and 5 for presentation of data from the Sonoma Valley Pinot Noir, Santa Maria Gewürztraminer, and Santa Maria Chardonnay trials, respectively.

[b] The Sonoma vineyard was infested with grape mealybug, and the Santa Maria vineyards were infested with obscure mealybug.