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Synthetic Pheromones as a management technique – Dispensers decrease *Linepithema humile* activity in a commercial vineyard

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ABSTRACT

BACKGROUND: Invasive ants, such as the Argentine ant, have often been reported to facilitate honeydew-producing hemipteran pests like mealybugs, which can be vectors of plant pathogens. Synthetic pheromones may offer a target-specific method to control such ants and consequently lower the abundance of honeydew producing pests. Here we report the results of a trial to suppress Argentine ants in grape vines using ant pheromone dispensers.

RESULTS: Compared with untreated controls, we observed a significant drop in Argentine ant activity on the ground, irrespective of whether pheromone dispensers were placed at ground level, within the canopy or in both locations. Ant counts in the canopy confirmed that Argentine ant abundance was reduced under the influence of the pheromone dispenser placed at ground level compared with untreated controls. However, placing dispensers only in the canopy did not reduce the numbers of ants within the canopy compared with untreated controls.

CONCLUSION: Our results showed that pheromone dispensers can significantly reduce Argentine ant foraging in grape vines, if they are positioned appropriately. This technique could potentially reduce the abundance of associated mealybugs and potentially attendant virus vectoring area wide.

1. INTRODUCTION

Invasive species can impose economic losses through decreased agricultural productivity and the associated costs of control measures (16, 21, 22). For instance, invasive ant species have often been reported to facilitate honeydew-producing hemipteran pests like mealybugs (10, 12), which can accelerate the distribution of plant pathogens like the virus causing *Mealybug wilt of pineapple* (11, 26) or the *Grapevine leafroll-associated virus 3* (GLRaV-3) (17, 35).

The invasive Argentine ant *Linepithema humile* M., native to South America, has successfully spread worldwide (10, 24, 28, 39). Its success has been partly attributed to its strong association with honeydew-producing insect species (27). In New Zealand, the invasive Argentine ant has been observed tending to mealybugs in horticultural crops, and is therefore regarded as likely to be a significant pest in a variety of crops, (14, 15) and has potential to spread significantly (36). Mealybugs are vectors of GLRaV-3 (8), an economically important disease of grapevines found throughout the world (5). Estimated costs range from $25000 to $40000 per hectare for different scenarios of yield reduction and quality penalties (1). Mealybug numbers on vine grapes could be significantly reduced simply by controlling Argentine ant numbers (20). Therefore, controlling these invasive ants is an important strategy to reduce the spread and impact of mealybugs and potentially, any associated plant pathogens.

While insecticides are often used for pest management, the reported negative impacts of insecticides on pollinators (3, 7, 38) and even potential facilitation of invasive species (2), makes this a less and less desirable management approach. Pheromones may offer a target-specific and potentially more environmentally friendly approach for integrated pest management (40) and have become more prevalent over the years in push-pull strategies (6). Pheromones usage can be tailored to specific usage, for instance on organic crops, if encapsulated in a dispenser (41) or for area-wide control if applied by aerial dispersion (4). More recently, pheromones have also been used to manipulate foraging of invasive ant species like *Monomorium pharaonis* (23) or the Argentine ant (19).

The synthetic pheromone (Z)-9-hexadecenal (Z9-16:Ald) disrupts Argentine ant trail following behaviour (19, 29–31). High pheromone concentrations prevent Argentine ants from establishing stable trails, thus limiting their ability to successfully forage and recruit greater numbers of con-specifics to resources in the area (30). Additionally, this treatment can also provide a competitive advantage for other resident ant species to increase their foraging, when directly competing with the Argentine ant for a specific food resource (37). Therefore, the potential exists for this synthetic trail pheromone to provide a management technique which if successful, could reduce the risk of disruption to mealybug biological control (25) and in doing so, reduce the risk of plant pathogen spread.

In previous trials on Argentine ants, a variety of different products have been used to deploy the trail pheromone in the field. Trail disruption had been previously demonstrated using a loaded canuba wax point source (29), a micro-encapsulated sprayable product (30) and pheromone diluted in ethanol (31). Nishisue et al. (2010) reported a successful long-term control of Argentine ants and reduced foraging activity using a rope dispenser product by Shin-Etsu Chemical, Tokyo (34). These dispensers can be permanently deployed on an ant-infested site and continuously distribute pheromone to their immediate surroundings, without it being washed away by rain. Nishisue *et al.* (2010) tested the pheromone rope dispensers in an urban garden environment and deployed them on ~40 cm poles. To our knowledge, however, a successful deployment of this technology in a commercial agricultural environment has not yet been demonstrated.

Mealybugs are primarily located in the canopy of vines. Argentine ants form trails to harvest honeydew and in so doing, the ants also effectively protect mealybugs from predators and parasitoids (18). Because the different spatial scale of agricultural crops allows for much more three-dimensional-movement than in the urban grass environment in the previous trial (19), the deployment of a fixed pheromone dispenser also has to take into account the effect of pheromone placement within the crop. While a dispenser on the ground, around the crop might decrease the ability of ants to form a trail towards these plants, the effect might not extend into the canopy of a larger plant or fruit tree. This scenario could still allow ants that reach the upper parts of the plants to tend to mealybugs. On the other hand, a dispenser in the canopy of fruit plants could decrease the ability of ants to locate mealybugs.

Here we report the results of a trial to suppress Argentine ants in an agricultural environment using pheromone dispensers deployed in two ways to enable inferences about foraging and its disruption in a three dimensional space. Our aim was to lower ant abundance in the vine canopy, where mealybugs occur and assess the most effective management approach to achieve this goal. We tested the effects of artificial Argentine ant pheromone on ant abundance when the rope dispenser was placed on the ground and/or in the canopy. Previous studies reported a limit to the range of the pheromone (29); therefore we predicted that, placing the dispenser near the ground would have the greatest impact on Argentine ant ground activity, while dispensers in the vine canopy would have a greater impact on Argentine ant numbers in the vine canopy.

2. EXPERIMENTAL METHODS

2.1 Study site

The study was conducted in the late summer to autumn (20 February to 1 May 2013) on a commercial vineyard in Hawke’s Bay (39°63’S, 176°81’E), which is a winegrowing region on the east coast of New Zealand’s North Island. The study block was planted in Cabernet Sauvignon vines in 2001. The trial was carried out within the block using an area of unplanted land sufficiently large enough to accommodate our experimental design. The entire Cabernet Sauvignon planted area had been colonised by the Argentine ant since at least 2009. The Argentine ant was abundant (Fig. 1A) and no other ant species could be found in the immediate trial area. Presence of the ant within the trial area was confirmed by preliminary pitfall trap sampling during early February 2013.

2.2 Experimental setup

We placed 60 potted Pinot Noir grapevines (approximately 1 year old) into plastic buckets (10 L) (Fig. 1B), which were buried under the irrigation lines in the unplanted vine rows. A single 10-mm diameter hole in each bucket c. 50 mm from its base allowed the outflow of excess water from the daily irrigation. A total of 48 vines (incorporating four treatments as described below) were set up in four rows (Fig. 1C); the distance between plants was 2.5 m. A random numbers table was used to assign treatments as follows (n = 12 plants per treatment): Ground (G): one dispenser was wrapped around a short pole at a maximum height of 10 cm above ground. The pole was placed next to the vine, allowing pheromone to disperse without physically obstructing/preventing ants from walking up the vine; Canopy (C): one dispenser wrapped around the vine at a height of approximately 50 cm above ground; Ground + Canopy (GC): one dispenser on the ground and one in the vine canopy as described above; Untreated internal control (UI): brown electrical wire visually resembling the pheromone dispensers was tied in the canopy of the untreated internal control vines to account for any potential disturbance effects of the physical presence of these dispensers. To check for any potential area-wide effect of the pheromone, which might influence the untreated internal control, an additional 12 plants were established in a separate area at a distance of c. 20 m away from the main experimental area as an external untreated control (UE). Since individual plants were randomly assigned to treatment groups, we measured the length of the longest cane of each plant, to confirm whether a comparable amount of ‘canopy space’ between treatment groups was available.

We used a rope pheromone dispensers supplied by Shin-Etsu (Shin-Etsu Chemical, Tokyo, Japan; see details in Tanaka et al. 2009). It contained approximately 375 mg of (Z)-9-hexadecanal per meter, therefore each 20 cm dispenser contained 75 mg of pheromone, which, under stable conditions, would release the pheromone at an estimated rate of 1.2 mg per day (calculation based on constant temperature/delivery rate over 2 months; the manufacturer asserts a 2-month lifespan based on data for moth disruption. Dispensers were initially applied to vines in the field on 20 February 2013 and replaced on 15 March 2013.

To provide a natural food source for Argentine ants on the potted vines, plants were inoculated with 10–15 citrophilus mealybugs (*Pseudococcus calceolariae*) growing on a single piece of seed potato and placed into a cotton mesh bag (15 × 8 cm) (Fig. 1D). The 2–4 mm diameter apertures throughout the fabric allowed mealybugs to naturally disperse onto the grapevine and to be picked up and tended by the Argentine ant. Before placement in the vineyard, mealybug colonies were reared on seed potatoes (cv. Karaka) in a laboratory (23°C ±1.0°C).

Argentine ants were observed to approach the potted plants almost immediately after they had been dug in, walking over the side of the bucket and onto the plants trunks. Within 30 min of vine deployment, ants were observed investigating the cotton pockets containing the seed potatoes inoculated with mealybugs (Fig. 1A). Furthermore, mealybugs were observed moving out of the mesh pockets and onto the vines (Fig. 1E) within the same time.

2.3 Ground assessments

To assess if pheromone treatments would decrease Argentine ant abundance on the ground, we used pitfall traps to examine ant abundance. These traps were placed in the ground next to each plant on warm, dry and sunny days for 48 h. In total, 23 traps were placed in the main experimental area and an additional 6 traps were positioned in the ground in the external control area. The traps were filled with approximately 30 ml of water and propylene glycol at a ratio of 4:1 and a drop of dishwashing liquid to break the surface tension. The initial trapping was conducted on 15 February before the experiment was established. Subsequent trappings were undertaken on 15 March, 8 and 29 April.

2.4 Canopy assessments

To assess the effect on Argentine ant foraging in the vine canopy, we undertook 1-min counts of Argentine ant activity on each vine per treatment. Visual searches encompassed all parts of the vine, going methodically from bottom to top (trunk, crown, canes and leaves) and counting every ant seen within the allotted timeframe. Counts were undertaken on six separate occasions (25 February, 8, 16 and 26 March, 8 and 23 April 2013) on warm, dry and sunny days.

2.5 Statistical analysis

Statistical analyses were performed in IBM SPSS Statistics 20.00. Count data of pitfall traps and canopy counts were square root transformed. The pitfall trap dataset failed Levenes test for homogeneity of variance, which was accounted for by using a repeated measures linear general model (ANOVA) with a Games Howell post hoc test (9) to analyse the data for differences between treatment groups over time. Canopy count data were analysed using a repeated measures linear general model (ANOVA) with a Fishers LSD post hoc test, after passing Levenes test for homogeneity. The results are interpreted with a degree of caution because both datasets failed Mauchly's test of Sphericity (pitfall trap Mauchly's W = 0.251 df = 5 Greenhouse – Geisser = 0.659; canopy counts Mauchly's W = 0.606 df = 14 Greenhouse – Geisser = 0.852). A general linear model (ANOVA) was used to compare the available canopy space between the different groups.

3. RESULTS

3.1 Initial conditions

Argentine ants had been observed to be very abundant within the experimental area in the four years prior to establishment of the experiment, and this was confirmed by our initial pitfall trapping. No significant difference (ANOVA: df = 4; F = 0.801; p = 0.530) was found for Argentine ant density in the initial assessment amongst the set-up locations (Fig. 2).

We compared the amount of available ‘canopy space’ within each treatment group by measuring the length of the longest cane of each plant (Mean 1.24 m ± 0.39m) and found no significant difference for this factor (ANOVA: df = 4; F = 0.691; p = 0.601), therefore it was not included in further analysis.

3.2 Ground assessments

Argentine ant density was found to be significantly different between the external control (UE) and all treatments (Tab. 1), with a strong reduction of traffic around the treated vines (Fig. 2). We found no statistical difference amongst treatments (Tab. 1). Furthermore, no difference in Argentine ant activity on the ground was detected between the external (UE) and the internal control (UI).

3.3 Canopy assessments

During the 1-min counts, Argentine ants were observed to be present on all the sentinel vines, independent of treatment (Fig. 3). However, we found significantly fewer ants were found on the ground (G) and ground plus canopy (GC) treatment on all assessment dates compared with either of the untreated controls (UI, UE) (Tab. 1). Most interestingly, we did not observe any significant difference in ant numbers in the vine canopy between the canopy treatment (C) and either of the untreated controls (UI, UE) at any point during this study (Tab. 1). As during ground assessments, there was no difference between the ground

(G) and ground plus canopy (GC) treatments. Furthermore, we found no difference for Argentine ant numbers in the canopy between the external (UE) and the internal control (UI).

4. DISCUSSION

Our study presents a successful demonstration that the application of trail pheromones can suppress Argentine ant activity in an agricultural environment. In our initial assessment, we found the Argentine ant evenly distributed across the experimental area. Therefore, it could be assumed that all the vines included in our experimental plot had an equal risk of ant incursions. The analysis of our data, however, suggests that Argentine ant activity was significantly reduced in some treatments and their access onto these vines decreased following the addition of pheromone dispensers.

We predicted that, placing the dispenser near the ground would have the most suppressive effect on Argentine ant ground activity. While we observed a significant drop in ant abundance compared with controls, which was similar to previous reports by Tanaka et al. (2008), there was no effect between the dispenser locations on the ground traffic. The number of workers on the ground decreased by 73 to 79% after the establishment of the trial compared with their initial abundance, and activity continued to stay at a similar level in following weeks. This result suggests that the pheromone filaments and meandering plumes lowered random ground traffic and scouting independently of the exact location of the dispensers. More interestingly, an increased number of dispensers in the ground plus canopy (GC) treatment did not result in a further reduction in ant density, which suggests that the pheromone quantity distributed by the dispensers during our experiment was sufficiently high to achieve trail disruption, and within expectations of what had been previously used in pheromone disruption experiments on ants (37). Since no difference in activity was found between our external and internal control, either in pitfall traps, or in 1-min counts, we assume that the impact of the pheromone was limited to the immediate area of the dispensers and reduced quickly with increased distance from the source. Indeed another study (29) has suggested that the impact on Argentine ants is of short duration and they recover quickly once outside the area of effect.

We expected pheromone dispensers placed in the vine canopy would have the strongest suppressive effect on Argentine ant numbers in the vine canopy. Ant counts in the canopy confirmed that Argentine ant abundance was reduced under the influence of the pheromone dispenser, with distinct differences depending on the positioning of the pheromone dispenser. Most surprisingly, dispensers placed in the canopy did not appear to have any effect on the ability of the Argentine ant to access the canopy and therefore the mealybugs located therein. These results indicated that ground-placed dispensers, close to the base of a crop or fruit plant, may be more effective for Argentine ant control than treatment of the canopy. It seems likely that this placement would restrict or at least limit ant access onto the whole plant, as long as no other physical access points allow access to upper parts of the plant. It seems reasonable to assume that at least one reason for the difference in effectiveness between ground (G), ground plus canopy (GC) and canopy only (C) treatments was the ability of Argentine ants to use visual cues for orientation. Therefore, a ground treatment may reduce the ability of Argentine ants to recruit a continuous trail towards the trunk of the plant. However, once access to the plant has been established, we speculate that ants could use visual cues and or sense of gravity (13) to navigate up and down the plant and around the branches, which could severely decrease the effectiveness of pheromone applications.

Pheromones have shown a growing potential for integrated pest management (40), can be tailored to crops/pests (41) and adapted to local or area wide application (4). In our study, ant traffic was significantly reduced in the canopy of ground pheromone treated vines, therefore we conclude that the treatments likely altered normal ant foraging behaviour sufficiently to impede their ability to find and protect mealybugs, which offer honeydew. If so, this might result in major advantages for pest management. Firstly, it may be beneficial for mealybugs' natural enemies, particularly parasitoids, as a previous study has suggested that mealybug biological control may be negatively influenced by the presence of the Argentine ant (18). Conversely, this could also severely limit Argentine ants spread, as they depend on the access to honeydew to produce large enough numbers of workers to overcome other ant species (27). However, the target concentration of trail pheromone required for suppression of an entire population on a large scale is yet unknown, and thus it is too early to consider the feasibility of this control method. Earlier studies using higher pheromone concentrations achieved up to 90% reduction in foraging (29, 30), measured as trail presence after application of microencapsulation (as opposed to discrete dispensers), which was higher than the current study. The fast rate of recovery of normal trail response of the ants which has been reported previously (31) suggests that any ants in clean air may be able to trail normally, which would allow for uninterrupted foraging outside our experimental trial.

5. CONCLUSIONS

Our study presents a successful demonstration that the application of trail pheromones can suppress Argentine ant activity in an agricultural environment, which may also reduce the numbers of mealybugs. Our results are consistent with previous observations (19, 32, 33). Trail pheromone disruption is a target-specific and potentially more environmentally friendly control technique than the current area-wide use of insecticides, which have been shown to negatively impact other beneficial insect species (38).

The next step in the process is to progress our experimental design to a large-scale level. In our future studies we plan to assess the extent to which pheromone dispensers can successfully impede Argentine ant recruitment on mature agricultural crops, and therefore reduce its ability to interact with and protect mealybugs.

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REFERENCES

1. Atallah SS, Gomez MI, Fuchs MF, Martinson TE. 2011. Economic impact of grapevine leafroll disease on vitis vinifera cv. cabernet franc in finger lakes vineyards of new york. *Am. J. Enol. Vitic.* 63(1):73–79

2. Barbieri RF, Lester PJ, Miller AS, Ryan KG. 2013. A neurotoxic pesticide changes the outcome of aggressive interactions between native and invasive ants. *Proc. R. Soc. B Biol. Sci.* 280(1772):20132157–20132157

3. Blacquière T, Smagghe G, Van Gestel CAM, Mommaerts V. 2012. Neonicotinoids in bees: a review on concentrations, side-effects and risk assessment. *Ecotoxicology*. 21(4):973–92

4. Brockerhoff EG, Suckling DM, Kimberley M, Richardson B, Coker G, et al. 2012. Aerial application of pheromones for mating disruption of an invasive moth as a potential eradication tool. *PLoS One*. 7(8):e43767

5. Charles J, Cohen D, Walker J. 2006. A review of the ecology of grapevine leafroll associated virus type 3 (glrav-3). *New Zeal. Plant Prot.* 59:330–37

6. Cook SM, Khan ZR, Pickett JA. 2007. The use of push-pull strategies in integrated pest management. *Annu. Rev. Entomol.* 52(1):375–400

7. Cresswell JE. 2011. A meta-analysis of experiments testing the effects of a neonicotinoid insecticide (imidacloprid) on honey bees. *Ecotoxicology*. 20(1):149–57

8. Daane KM, Almeida RPP, Bell VA, Walker JTS, Botton M, et al. 2012. Biology and management of mealybugs in vineyards. In *Arthropod management in Vineyards*, ed NJ Bostanian, C Vincent, R Isaacs, pp. 271–307. Dordrecht: Springer Netherlands

9. Games PA, Howell JF. 1976. Pairwise multiple comparison procedures with unequal n's and/or variances: a monte carlo study. *J. Educ. Behav. Stat.* 1(2):113–25

10. Holway D a., Lach L, Suarez A V., Tsutsui ND, Case TJ. 2002. The causes and consequences of ant invasions. *Annu. Rev. Ecol. Syst.* 33(1):181–233

11. Jahn GC, Beardsley JW, Gonzalez-Hernandez H. 2003. A review of the association of ants with mealybug wilt disease of pineapple. *Proc. Hawaiian Entomol. Soc.* 36:9–28

12. Johnson C, Agosti D, Delabie JH, Dumpert K, Williams DJ, et al. 2001. Acropyga and azteca ants (Hymenoptera: Formicidae) with scale insects (Sternorrhyncha: Coccoidea): 20 million years of intimate symbiosis. *Am. Museum Novit.* 3335(3335):1–18

13. Khuong A, Lecheval V, Fournier R, Blanco S, Weitz S, et al. 2013. How do ants make sense of gravity? a boltzmann walker analysis of lasius niger trajectories on various inclines. *PLoS One*. 8(10):e76531

14. Lester PJ, Baring CW, Longson CG, Hartley S. 2003. Argentine and other ants (Hymenoptera: Formicidae) in new zealand horticultural ecosystems: distribution, hemipteran hosts, and review. *New Zeal. Entomol.* 26:79–89

15. Lester PJ, Longson CG. 2002. Argentine ant distribution investigation: horticultural areas. Report to MAF Biosecurity, Victoria University of Wellington

16. Mack RN, Simberloff D, Mark Lonsdale W, Evans H, Clout M, Bazzaz FA. 2000. Biotic invasions: causes, epidemiology, global consequences, and control. *Ecol. Appl.* 10(3):689–710

17. Maree HJ, Almeida RPP, Bester R, Chooi KM, Cohen D, et al. 2013. Grapevine leafroll-associated virus 3. *Front. Microbiol.* 4:82

18. Mgocheki N, Addison P. 2010. Spatial distribution of ants (Hymenoptera: Formicidae), vine mealybugs and mealybug parasitoids in vineyards. *J. Appl. Entomol.* 134(4):285–95

19. Nishisue K, Sunamura E, Tanaka Y, Sakamoto H, Suzuki S, et al. 2010. Long-term field trial to control the invasive argentine ant (Hymenoptera: Formicidae) with synthetic trail pheromone. *J. Econ. Entomol.* 103(5):1784–89

20. Phillips PA, Sherk CJ. 1991. To control mealybugs, stop honeydew-seeking ants. *Calif. Agric.* 45(2):2–4

21. Pimentel D, Lach L, Zuniga R, Morrison D. 2000. Enviromental and economic costs associated with nonindigenous species in the united states. *Bioscience*. 50(1):53–65

22. Pimentel D, Zuniga R, Morrison D. 2005. Update on the environmental and economic costs associated with alien-invasive species in the united states

23. Robinson EJH, Jackson DE, Holcombe M, Ratnieks FLW. 2005. Insect communication: “no entry” signal in ant foraging. *Nature*. 438(7067):442

24. Roura-Pascual N, Suarez A V, McNyset K, Gómez C, Pons P, et al. 2006. Niche differentiation and fine-scale projections for argentine ants based on remotely sensed data. *Ecol. Appl.* 16(5):1832–41

25. Samways M, Nel M, Prins A. 1982. Ants (Hymenoptera: Formicidae) foraging in citrus trees and attending honeydew-producing homoptera. *Phytophylactica*. 14(4):155–57

26. Sether DM, Melzer MJ, Busto J, Zee F, Hu JS. 2005. Diversity and mealybug transmissibility of ampeloviruses in pineapple. *Plant Dis.* 89(5):450–56

27. Shik JZ, Silverman J. 2013. Towards a nutritional ecology of invasive establishment: aphid mutualists provide better fuel for incipient argentine ant colonies than insect prey. *Biol. Invasions*. 15(4):829–36

28. Suarez A V, Holway DA, Case TJ. 2001. Patterns of spread in biological invasions dominated by long-distance jump dispersal: insights from argentine ants. *Proc. Natl. Acad. Sci. U. S. A.* 98(3):1095–1100

29. Suckling DM, Peck RW, Manning LM, Stringer LD, Cappadonna J, El-Sayed AM. 2008. Pheromone disruption of argentine ant trail integrity. *J. Chem. Ecol.* 34(12):1602–9

30. Suckling DM, Peck RW, Stringer LD, Snook K, Banko PC. 2010. Trail pheromone disruption of argentine ant trail formation and foraging. *J. Chem. Ecol.* 36(1):122–28

31. Suckling DM, Stringer LD, Corn JE. 2011. Argentine ant trail pheromone disruption is mediated by trail concentration. *J. Chem. Ecol.* 37(10):1143–49

32. Tanaka Y, Nishisue K, Sunamura E. 2009. Trail-following disruption in the invasive argentine ant with a synthetic trail pheromone component (z)-9-hexadecenal. *Sociobiology*. 1:139–52

33. Tanaka Y, Sunamura E, Nishisue K, Terayama M, Sakamoto H, et al. 2008. Response of the invasive argentine ant to high concentration of a synthetic trail pheromone component suggests a potential control strategy of pest ants. *Ari*. 31:43–50

34. Tatsuki S, Terayama M, in- ventors; Shin-Etsu Chemical, Co., Ltd. A. 2005. Behavior-disrupting agent and behavior-disrupting method of argentine ant. *U.S. Pat. 0209344 A1*. 2(12):6–9

35. Tsai C-W, Chau J, Fernandez L, Bosco D, Daane KM, Almeida RPP. 2008. Transmission of grapevine leafroll-associated virus 3 by the vine mealybug (*Planococcus ficus*). *Phytopathology*. 98(10):1093–98

36. Ward DF, Green C, Harris RJ, Hartley S, Lester PJ, et al. 2010. Twenty years of argentine ants in new zealand: past research and future priorities for applied management. *New Zeal. Entomol.* 33(1):68–78

37. Westermann FL, Suckling DM, Lester PJ. 2014. Disruption of foraging by a dominant invasive species to decrease its competitive ability. *PLoS One*. 9(3):e90173

38. Whitehorn PR, O’Connor S, Wackers FL, Goulson D. 2012. Neonicotinoid pesticide reduces bumble bee colony growth and queen production. *Science*. 336(6079):351–52

39. Wild AL. 2004. Taxonomy and distribution of the argentine ant, linepithema humile (hymenoptera: formicidae). *Ann. Entomol. Soc. Am.* 97(6):1204–15

40. Witzgall P, Kirsch P, Cork A. 2010. Sex pheromones and their impact on pest management. *J. Chem. Ecol.* 36(1):80–100

41. Zehnder G, Gurr GM, Kühne S, Wade MR, Wratten SD, Wyss E. 2007. Arthropod pest management in organic crops. *Annu. Rev. Entomol.* 52(July):57–80

Table 1. Mean difference, standard error, p-values and confidence intervals between treatments from the repeated measures linear model for the ground assessment (Games Howell post hoc test) and canopy assessment (Fishers LSD post hoc test). C = canopy; G = ground; GC = ground plus canopy; UI = untreated internal control; UE = untreated external control. Statistically significant differences between treatments during each assessment are denoted by one asterix (α = 0.05).

						95% Confidence Interval	
			Mean Difference (I-J)	Std. Error	Sig.	Lower Bound	Upper Bound
Ground assessment (pitfall traps)	UE	C	2.858*	.562	.001	1.073	4.643
		G	2.786*	.535	.001	1.060	4.512
		GC	3.201*	.595	.000	1.331	5.070
		UI	1.051	.811	.868	-1.528	3.630
	C	G	-.0721	.390	1.000	-1.287	1.143
		GC	.342	.469	.997	-1.122	1.808
		UE	-2.858*	.562	.001	-4.643	-1.073
		UI	-1.807	.723	.206	-4.204	.589
Canopy assessment (1-min counts)	UE	C	-.082	.4907	.868	-1.065	.901
		G	1.447*	.4907	.005	.464	2.431
		GC	1.108*	.4907	.028	.124	2.091
		UI	.592	.4907	.233	-.391	1.575
	C	G	1.529*	.4907	.003	.546	2.513
		GC	1.190*	.4907	.019	.206	2.173
		UE	.082	.4907	.868	-.901	1.065
		UI	.673	.4907	.175	-.309	1.657

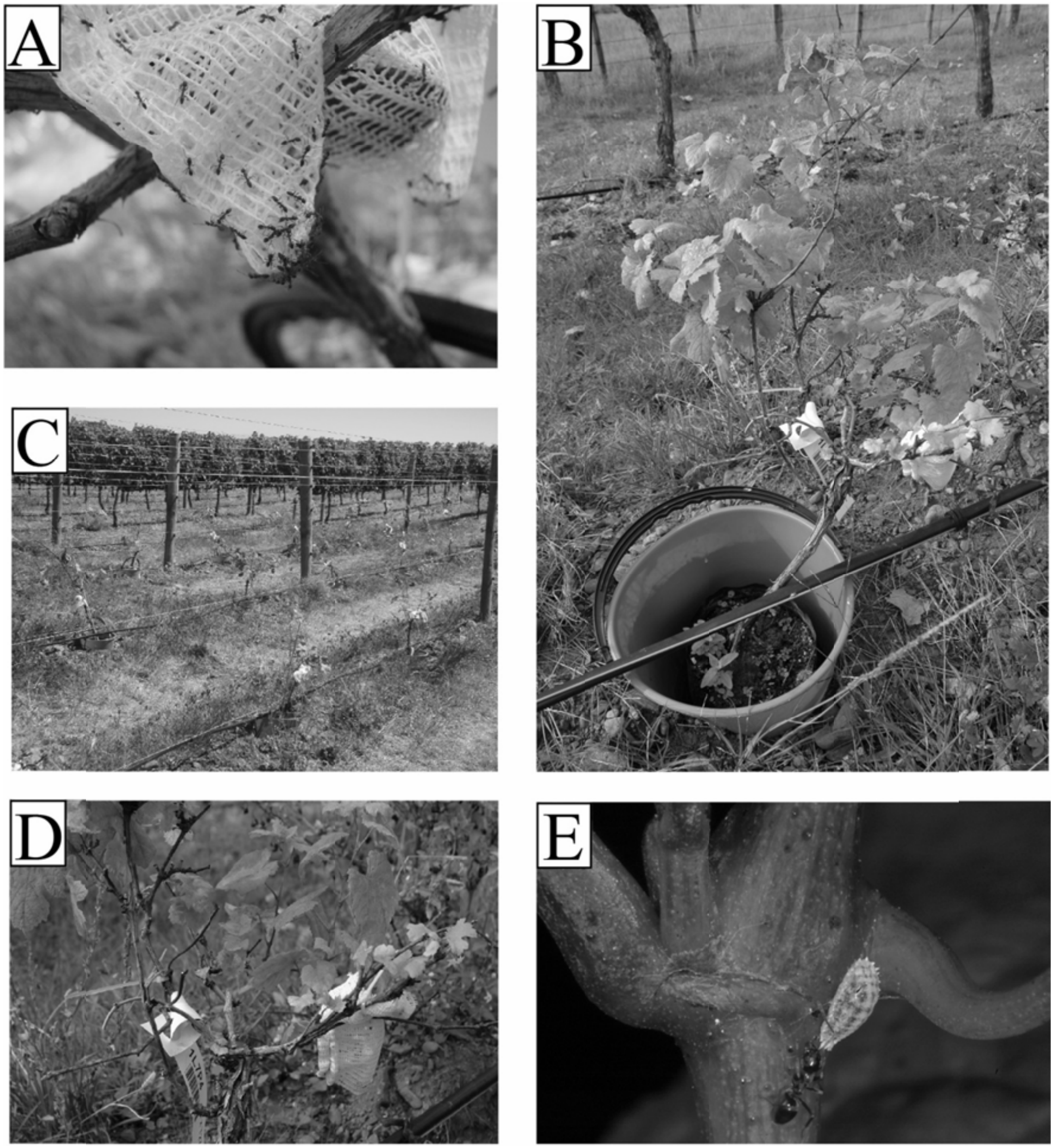

Figure 1. A) Argentine ants approaching mesh pockets with mealybugs in vine canopy before placement of pheromone dispensers. B) Experimental setup of a single grapevine, placed in a bucket under the irrigation line. C) Experimental setup of the trial. D) A close-up of the canopy treatment with pheromone disperser (brown wire) attached to the vine (left) and white mesh pocket containing mealybugs on a segment of seed potato (right). E) Argentine ant tending to a citrophilus mealybug on one of the experimental plants.

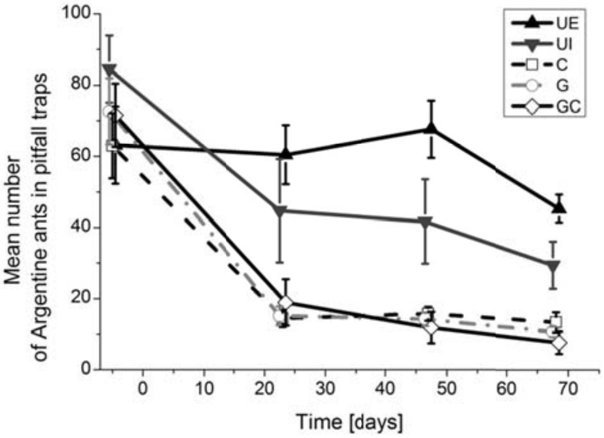

Figure 2. Mean (± SEM) numbers of Argentine ants recorded in pitfall traps established in the Hawke's Bay vineyard study block. Assessments were undertaken between 15 February (before pheromone treatment) and 29 April 2013 (n = 12 pitfall traps next to G; n = 11 pitfall traps next to UE, C and GC; n = 10 pitfall traps next to UI). C = canopy; G = ground; GC = ground plus canopy; UI = untreated internal control; UE = untreated external control.

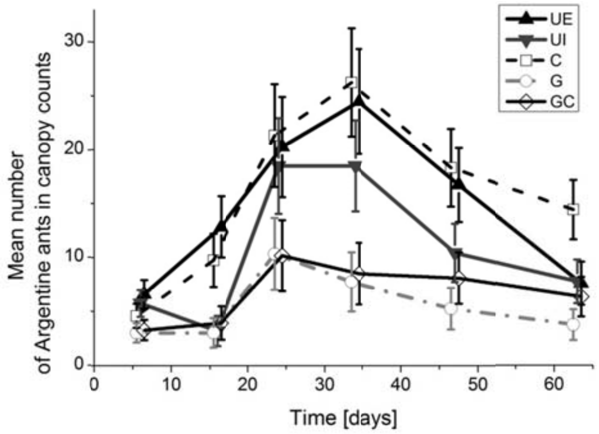

Figure 3. Mean (± SEM) numbers of Argentine ants recorded during 1-min counts in the Hawke's Bay vineyard study block (n = 12 mealybug-inoculated Pinot Noir grapevines per treatment). C = canopy; G = ground; GC = ground plus canopy; UI = untreated internal control; UE = untreated external control.