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Trunk injection of systemic insecticides to control stem and leaf gall wasps, *Josephiella* species (Hymenoptera: Agaonidae), on Chinese banyan (Rosales: Moraceae) in Hawaii

*Bishnu P. Bhandari and Zhiqiang Cheng**

Abstract

Chinese banyan, *Ficus microcarpa* L. f. (Rosales: Moraceae), is a popular landscape tree in many tropical regions of the world. In Hawaii, these trees are severely infested by 2 host-specific insect species in the family Agaonidae (Hymenoptera: Chalcidoidea): the Chinese banyan leaf gall wasp, *Josephiella microcarpae* Beardsley & Rasplus, and the stem gall wasp *Josephiella* sp. (currently being described). Infestations by these insects result in gall formation on young leaves and shoots, premature leaf drop, new shoot death, poor tree health, and eventually death of the tree. We evaluated the efficacy and persistence of 2 systemic insecticides, imidacloprid and emamectin benzoate, with or without phosphorous acid amendment, delivered through trunk injection to control these 2 wasp species in Honolulu, Hawaii. Although both systemic insecticides had some effect against leaf gall wasps for up to 18 mo post treatment, only emamectin benzoate persisted against stem gall wasps for up to 14 mo post treatment. Phosphorous acid amendment did not provide any benefits for Chinese banyans to mitigate wasp infestations. In conclusion, trunk injection of emamectin benzoate could be a feasible management strategy to control stem and leaf gall wasps on Chinese banyans in Hawaii.

Key Words: agaonid wasp; imidacloprid; emamectin benzoate

Resumen

El laurel de indias, *Ficus microcarpa* L. f. (Rosales: Moraceae), es un árbol popular del campo en muchas regiones tropicales del mundo. En Hawai, estos árboles están severamente infestados por 2 especies de insectos específicos de la familia Agaonidae (Hymenoptera: Chalcidoidea): la avispa de agallas de hojas del laurel de indias, *Josephiella microcarpae* Beardsley y Rasplus, y la avispa de agallas del tallo *Josephiella* sp. (que se describe en la actualidad). Las infestaciones por estos insectos resultan en la formación de agallas en las hojas y los brotes jóvenes, la caída prematura de las hojas, la muerte de los nuevo brotes, la mala salud del árbol, y finalmente, la muerte del árbol. Se evaluó la eficacia y persistencia de 2 insecticidas sistémicos, imidacloprid y benzoato de emamectina, con o sin la adición de ácido fosforoso, entregados a través de inyección en el tronco para controlar estas 2 especies de avispas en Honolulu, Hawaii. Aunque ambos insecticidas sistémicos tuvieron algún efecto contra las avispas de las agallas de hojas por hasta 18 meses después del tratamiento, sólo el benzoato de emamectina persistia contra las avispas de las agallas del tallo por hasta 14 meses pos-tratamiento. La adición del ácido fosforoso no proveyó ningún beneficio para los árboles del laurel de indias para mitigar las infestaciones de avispas. En conclusión, la inyección en el tronco de benzoato de emamectina podría ser una estrategia de manejo viable para controlar las avispas de las agallas de las hojas y de los tallos en los árboles del laurel de indias en Hawaii.

Palabras Clave: avispa agaonid; imidacloprid; benzoato de emamectina

Chinese banyan, *Ficus microcarpa* L. f. (Rosales: Moraceae), is native to diverse geographical locations ranging from Ceylon to India, southern China, Ryukyu Islands, Australia, and New Caledonia (Wagner et al. 1999; Starr et al. 2003). This tree is widespread and commonly found in landscapes and as a container tree in many tropical regions of the world. It has some other common names, most notably Cuban laurel and Indian laurel. It is a popular landscape tree in Hawaii, where it was introduced in 1921 (Ramirez & Montero 1988). Chinese banyan is pollinated by *Eupristina verticillata* Waterston (Hymenoptera: Agaonidae), which was purposely introduced to Hawaii in 1938 (Pemberton 1939).

The non-pollinating agaonid fig wasp, *Josephiella microcarpae* Beardsley & Rasplus (Hymenoptera: Agaonidae), which forms galls on the leaves of *F. microcarpa*, was first found in Hawaii in 1989 (Starr et al. 2003), and the species was described in 2001 (Beardsley & Rasplus 2001). This wasp was also discovered in California in 1997 (Anynomous 1998), and in Florida in 2007 (Caldwell 2008). It remains as a pest problem in certain areas of Florida (Caldwell 2008). The adult leaf gall wasps are dark brown, with pale yellow appendages, and about 2.2 mm long (female). The adult female wasps deposit eggs in very young terminal leaves, but not mature leaves, and insert multiple eggs. As a larva develops, the leaf tissue swells around it and forms the gall. Larvae pupate inside galled leaves both on the tree and on the soil surface. When the adult emerges, it chews a round hole on the lower portion of the leaf and exits. The male wasps are similar to females except that they are smaller in size (about 1.1 mm) and have shorter antennae (7 segments) than females (8 segments) (Beardsley & Rasplus 2001).

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In Jul 2012, another non-pollinating agaonid fig wasp, *Josephiella* sp. (Hymenoptera: Agaonidae), was discovered in Honolulu (HDOA 2012). This pest wasp induces galls on young branches, resulting in defoliation and ultimately death of young shoots. The characteristics and mode of damage of the newly discovered stem gall wasp are similar to those of the leaf gall wasp; however, stem gall wasps forming galls on the young twigs are more damaging to tree health than leaf gall wasps. The stem gall wasp is an undescribed species and has been discovered only in Hawaii to date. Our laboratory and field observations suggest that the leaf gall wasps typically have a 3 to 4 mo life cycle, whereas the stem gall wasps typically take 5 to 6 mo to complete the life cycle. We have not observed any natural enemy activities against these wasp species to date. A systemic insecticide treatment could be a viable management tactic at this stage.

Imidacloprid and emamectin benzoate are 2 systemic insecticides known to have some effect against gall-forming insects (Doccola et al. 2009). Imidacloprid, a neonicotinoid, moves through plant tissues after application and disturbs nerve impulse transmission, resulting in death (Fossen 2006). Emamectin benzoate is derived from abamectin, a natural fermentation product of the soil bacterium *Streptomyces avermitilis* (ex Burg et al.) Kim and Goodfellow (Actinomycetales: Streptomycetaceae). Its mode of action includes binding of insect neuron cells, cessation of cell function, and ultimately disruption of nerve impulses resulting in paralysis and death (USEPA 2009). The primary route of toxicity for both insecticides is through ingestion. Phosphorous acid, although mainly used as fungicide, could possibly have some positive effect by stimulating the plant natural defense system and overall health (Smillie et al. 1989) and was thus included in this study. Because Chinese banyan is pollinated only by *E. verticillata*, negative impacts of imidacloprid and emamectin benzoate in this plant–pest dynamic on other common pollinators such as honey bees are unlikely.

Trunk injection can be used to treat trees to combat problems related to insects, diseases, and nutrient deficiencies. This method typically requires less active ingredient as compared with conventional methods such as foliar sprays and soil drenching (Norris 1965; Doccola & Wild 2012). A trunk injection system is relatively simple and quick, and the chemicals can take only a few hours to weeks to reach all parts of trees, depending on the specific insecticides and tree characteristics (Doccola et al. 2009). The difference in flow rate among chemicals could be due to the dilution of the chemical formulation, tree canopy, tree size, and tree health (McWain & Gregory 1973; Doccola et al. 2009). Unlike conventional methods, trunk injection systems directly inject chemicals into the vascular system of the tree without exposing the environment to the chemicals.

The overall objective of this study was to determine the efficacy and persistence of 2 systemic insecticides, imidacloprid and emamectin benzoate, with or without phosphorous acid amendment and delivered through trunk injection to control stem and leaf gall wasps on Chinese banyan. We hypothesized that 1) imidacloprid and emamectin benzoate would be effective against stem and leaf gall wasps; 2) effectiveness would last for 2 yr; and 3) phosphorous acid amendment would help suppress gall wasps by enhancing the plant's natural defense system and overall health.

Materials and Methods

EXPERIMENTAL DESIGN

This research was conducted on the campus of the University of Hawaii at Manoa (21.2970°N, 157.8170°W) in Honolulu, Hawaii. We included 45 Chinese banyan trees, *F. microcarpa*, in this research. Most trees included were assumed to be of similar age based on their diameter at breast height (DBH), although their exact ages were uncertain. These trees were all infested with both stem and leaf gall wasps at similar levels of infestation. The mean DBH of trees was 121.7 cm ranging from 53 cm to 231 cm. There were 5 treatments: imidacloprid (5%) at 8 mL per 2.54 cm DBH, emamectin benzoate (4%) at 10 mL per 2.54 cm DBH, imidacloprid (5%) at 8 mL per 2.54 cm DBH + phosphorous acid (45.8%) at 7 mL per 2.54 cm DBH, emamectin benzoate (4%) at 10 mL per 2.54 cm DBH + phosphorous acid (45.8%) at 7 mL per 2.54 cm DBH, and untreated control. There were 9 trees per treatment. The Arborjet QUIK-jet micro infusion system and pressurized Tree IV system were used for injections in Jul 2013. Trunk injection equipment and chemicals tested were the products of Arborjet, Inc. (Woburn, Massachusetts). The trade names of these chemicals are TREE-age (emamectin benzoate), IMA-jet (imidacloprid), and PHOSPHO-jet (phosphorous acid).

DATA COLLECTION

Sampling was initiated 3 mo after injection, assuming the time required for chemical distribution throughout trees and the approximate time required for completing the life cycles by both stem and leaf gall wasp species, as treatments were intended to reduce gall formation on new leaves and shoots, rather than acting as a curative treatment for previously infested plant material.

Three new terminal shoots, each about 45 cm in length, were collected monthly from 3 to 12 mo after treatment (Oct 2013 to Jul 2014), and then bi-monthly from 14 to 22 mo post treatment (Sep 2014 to May 2015), from the tree canopy by using a 10 m long tree pruner. Shoots were collected from 3 sub-trunks of various sizes taken from each tree to ensure the samples were representative. Shoots with old exit holes were not considered as current infestation because the wasps had already completed their life cycle and exited from the holes. Seasonal variation in infestations of both leaf and stem gall wasps was limited, as the wasps were consistently active throughout the year in Honolulu, Hawaii.

These samples were used to determine: number of stem galls per 45 cm length, stem infestation level (1–5, 1 being no infestation and 5 being severely infested, i.e., the shoot fully covered with stem galls), percentage of leaves infested, and leaf infestation level (1–5, 1 being no infestation and 5 being severely infested, i.e., multiple galls on each leaf and most of the leaves infested on shoot). In the 14th and 22nd months after treatment, visual ratings on new shoot emergence and overall tree health were evaluated. The ratings were defined as follows: for tree condition, 1 = excellent, 2 = good, 3 = fair, 4 = poor, 5 = dead; for new shoot emergence, 1 = many, 2 = moderate, 3 = some, 4 = few, 5 = very few. Excellent tree health condition means the tree is in good shape with an abundance of new shoot growth and no dead shoots or branches.

STATISTICAL ANALYSES

Statistical analyses were conducted using MINITAB (Version 15, Minitab, Inc., State College, Pennsylvania), and differences were considered significant at $P \leq 0.05$. For number of stem galls and percentage of leaves infested, 1-way analysis of variance (ANOVA) was performed, with treatment and time as the main factors. Tukey's tests were conducted for pairwise comparison. For all other variables measured, non-parametric Kruskal–Wallis tests were conducted for treatment effect at each sampling time, and pairwise comparison was conducted using Dunn's method.

Results

Phosphorous acid was expected to have some effect against gall wasps through enhanced tree health and enhanced natural defense

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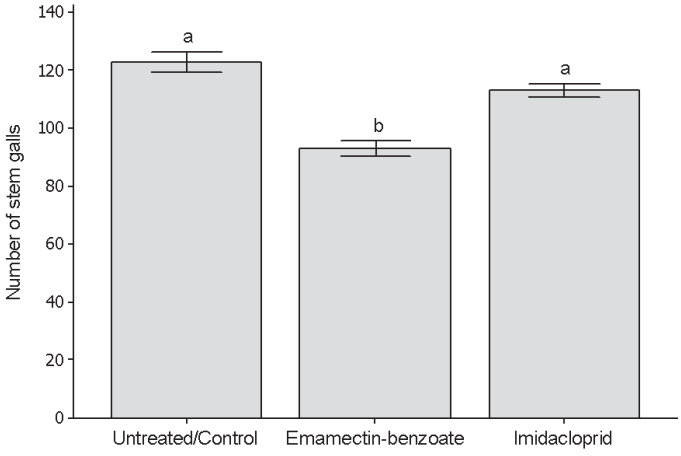


Fig. 1. Mean number (± SE) of stem galls (on 45 cm shoots) associated with chemical treatments and untreated control. Means with the same letter are not significantly different (ANOVA).

in combination with systemic insecticides. However, the data did not show any benefit of a phosphorous acid amendment to trees against wasps. Therefore, we combined treatments with and without phosphorous acid and analyzed the data from the 2 insecticide treatments (imidacloprid and emamectin benzoate) and the untreated control. The interaction of treatment and month after treatment was not significantly different among treatments in terms of number of stem galls (ANOVA, $F = 1.47$; df = 28; $P = 0.055$) and percentage of leaves infested (ANOVA, $F = 1.4$; df = 28; $P = 0.080$). So, for these 2 parameters, the main treatment effect was presented in Figs. 1 and 3.

Emamectin benzoate was significantly more effective than imidacloprid in suppressing stem gall formation (Fig. 1). Starting from the 4th mo after treatment, the trend indicated decreased stem gall infestation levels in trees treated with emamectin benzoate as compared with trees treated with imidacloprid and untreated control trees. The trend was consistent for up to 14 mo after treatment (Fig. 2). We observed up to a 46% reduction in the number of new stem galls in trees treated with emamectin benzoate as compared with the untreated controls. Results did not indicate significant differences for stem infestation levels after 14 mo post treatment.

Emamectin benzoate was effective at reducing leaf gall formation relative to both the imidacloprid treatment and the untreated control (Fig. 3). However, both insecticides were observed to be effective against leaf gall wasps, with significant reduction in gall formation on leaves of new shoots. With emamectin benzoate, significant effects were observed starting from the 4th mo after treatment, and this pat-

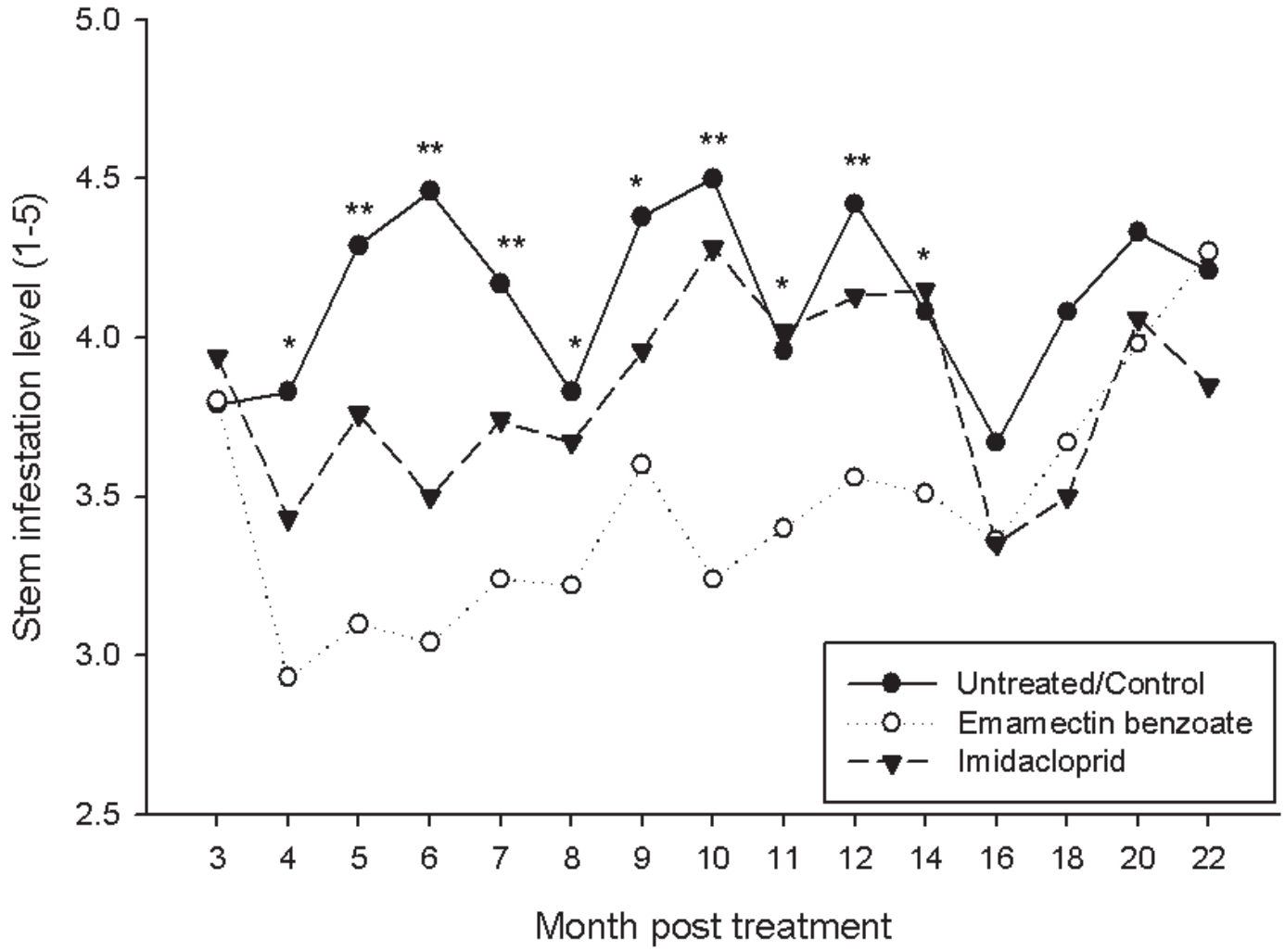


Fig. 2. Mean stem gall infestation levels on new shoots associated with chemical treatments and untreated control (1–5, where 1 = no infestation and 5 = severe infestation), where * indicates $P \leq 0.05$ and ** indicates $P \leq 0.01$ within each sampling month (Kruskal–Wallis test).

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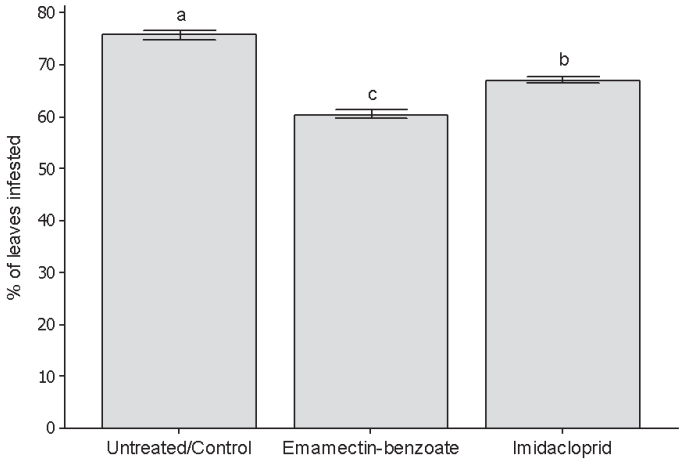


Fig. 3. Mean percentage (± SE) of leaves infested with leaf gall wasps associated with chemical treatments and untreated control. Means with the same letter are not significantly different (ANOVA).

tern persisted for up to 18 mo after treatment (Fig. 4). The reduction in mean percentage of leaves infested on trees treated with emamectin benzoate was up to 31% as compared with the untreated controls.

Evaluations of overall tree health and new shoot emergence were conducted in Sep 2014 and May 2015, 14 and 22 mo after treatments, respectively. Based on the results at 14 mo after treatment (Fig. 5), trees treated with emamectin benzoate had a significantly better tree health condition rating compared with trees treated with imidacloprid and untreated control trees (Kruskal–Wallis test, H = 8.18; df = 2; P = 0.017). Trees treated with both insecticides produced significantly more new shoots as compared with the untreated trees (Kruskal–Wallis test, H = 11.32; df = 2; P = 0.003).

In the 22nd mo after treatment, a similar pattern was observed on the rating of tree health condition, with significant differences between treatments and untreated control (Kruskal–Wallis test, H = 7.92; df = 2; P = 0.019). However, the rating on new shoot emergence was not significant (Kruskal–Wallis test, H = 4.11; df = 2; P = 0.128) among the trees under either treatment or the untreated control trees (Fig. 6).

Discussion

The key approach to protect Chinese banyans from infestation with stem and leaf gall wasps is to prevent and reduce new infestations. Despite severe stem gall wasp pre-treatment infestations, trees treated with emamectin benzoate maintained canopy with reduced new

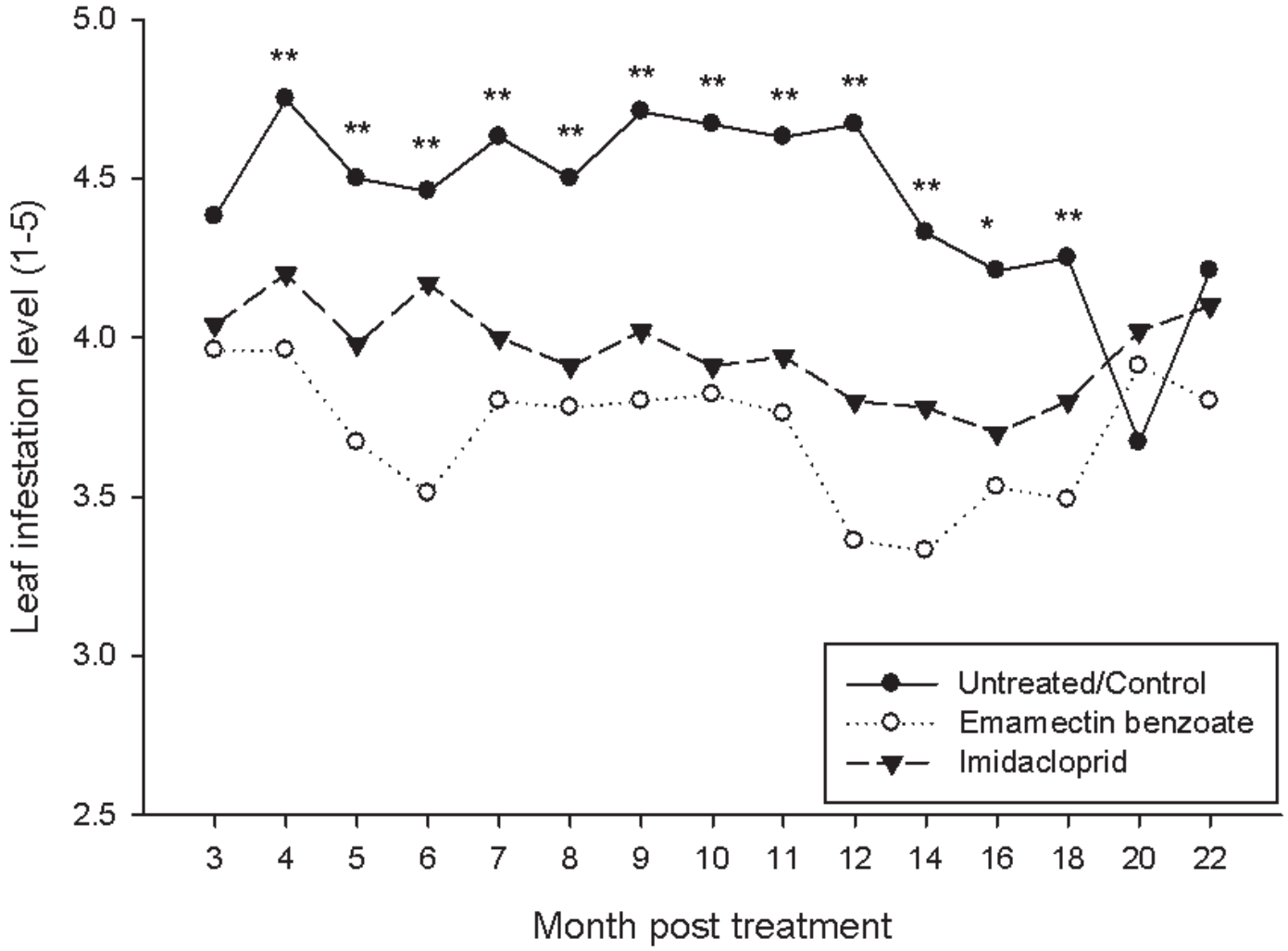


Fig. 4. Mean leaf gall infestation level on new shoots associated with chemical treatments and untreated control (1–5, where 1 = no infestation and 5 = severe infestation), where * indicates $P \leq 0.05$ and ** indicates $P \leq 0.01$ within each sampling month (Kruskal–Wallis test).

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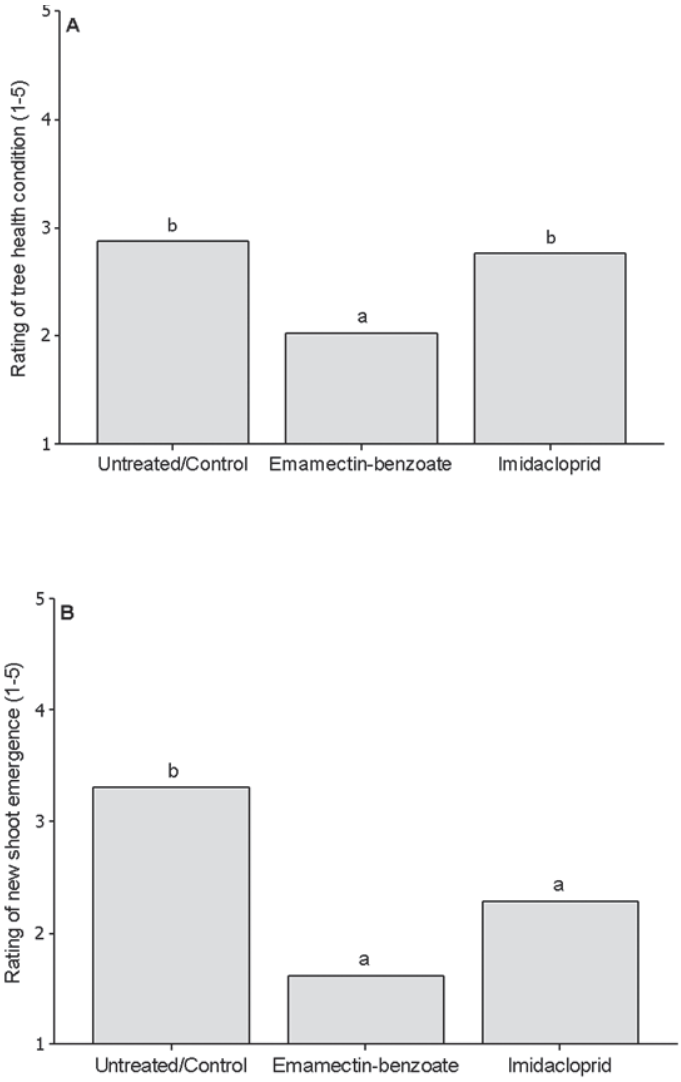


Fig. 5. Mean ratings of tree health condition (A) and mean ratings of new shoot emergence (B) 14 mo after treatment (rating of tree health condition: 1 = excellent, 2 = good, 3 = fair, 4 = poor, 5 = dead; rating of new shoots emergence: 1 = many, 2 = moderate, 3 = some, 4 = few, 5 = very few). Means with the same letter are not significantly different (Kruskal–Wallis test).

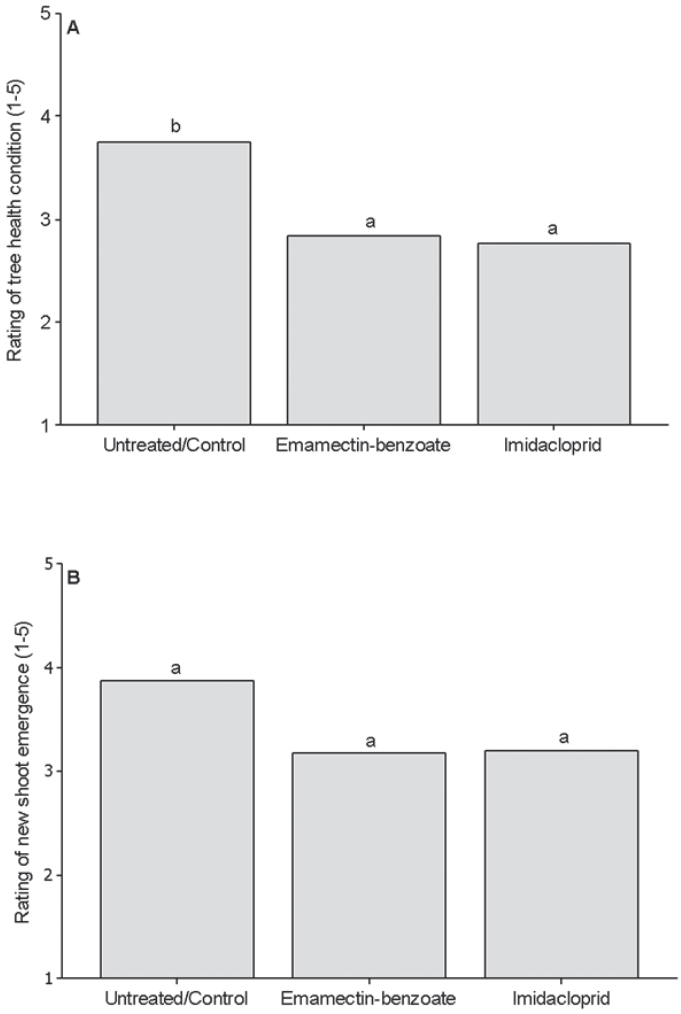


Fig. 6. Mean ratings of tree health condition (A) and mean ratings of new shoot emergence (B) 22 mo after treatment (rating of tree health condition: 1 = excellent, 2 = good, 3 = fair, 4 = poor, 5 = dead; rating of new shoots emergence: 1 = many, 2 = moderate, 3 = some, 4 = few, 5 = very few). Means with the same letter are not significantly different (Kruskal–Wallis test).

gall formation on leaves and stems, and were protected from further canopy loss attributable to gall wasps.

Trunk injection systems have shown variable results of effectiveness against insect pests. Previous studies have shown that trunk-injected emamectin benzoate provided excellent activity against some insect pests, such as the emerald ash borer, *Agrilus planipennis* Fairmaire (Coleoptera: Buprestidae) (Herms et al. 2009; Smitley et al. 2010; McCullough et al. 2011), the eastern tent caterpillar, *Malocosoma americanum* F. (Lepidoptera: Lasiocampidae) (Potter et al. 2005), and southern pine engravers, *Ips* species (Coleoptera: Curculionidae) (Grosman & Upton 2006). Smitley et al. (2010) found nearly 100% control of emerald ash borer larvae, *A. planipennis*, on green ash trees treated with trunk-injected emamectin benzoate at the rate of 0.1 to 0.4 gm active ingredient per inch DBH. The trunk-injected emamectin benzoate was highly effective against adults and larvae of the emerald ash borer for 2 seasons after treatment, but the trunk-injected imidacloprid did not have an effect against ash borer larvae (Herms et al. 2009; McCullough et al. 2011). A study on trunk injection of the insecticides abamectin and imidacloprid using the Mauget system (JJ Mauget Company Inc., Los Angeles, California) to manage the gall wasps *Callirhytis cornigera* (Osten Sacken) (Hymenoptera: Cynipidae) on pin oak, *Quercus palustris* Münchh. (Fagales: Fagaceae), was conducted by Eliason & Potter (2000). They found abamectin (chemically similar to emamectin benzoate) to be effective against leaf gall wasps. Consistent with these studies, our study showed that emamectin benzoate effectively suppressed stem and leaf gall wasps on Chinese banyans for at least 14 mo and 18 mo, respectively, after tree injection application. We observed up to 46% reduction in stem gall formation on trees treated with emamectin benzoate.

A study to investigate the effectiveness of emamectin benzoate and imidacloprid against erythrina gall wasps, *Quadrastichus erythrinae* Kim (Hymenoptera: Eulophidae), on Wiliwili trees, *Erythrina sandwicensis* Degener (Fabales: Fabaceae), indicated some effect of emamectin benzoate against erythrina gall wasps, but overall it was less effective than imidacloprid (Doccola et al. 2009). In contrast, our findings suggest that emamectin benzoate is more effective than imidacloprid against banyan stem gall wasps and that imidacloprid does not have significant effects against stem gall wasps on Chinese banyans.

Imidacloprid has been used in soil and trunk injection systems to manage a broad range of insect pests, and it has been found to be most

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effective against hemipteran pests. Trunk injection of imidacloprid was conducted using the Mauget system on Eastern hemlocks, *Tsuga canadensis* (L.) Carrière (Pinales: Pinaceae), to determine its effectiveness on the hemlock woolly adelgid, *Adelges tsugae* Annand (Aphidomorpha: Adelgidae) (Eisenback et al. 2014). The researchers found that imidacloprid reduced *A. tsugae* populations after 3 and 4 yr, resulting in new shoot growth and better visual ratings of tree health. Young (2002) studied the effectiveness of trunk-injected imidacloprid utilizing the J. J. Mauget system to manage the red gum lerp psyllid, *Glycaspis brimblecombei* Moore (Hemiptera: Aphalaridae), on red gum eucalyptus trees, *Eucalyptus camaldulensis* Dehnhardt (Myrtales: Myrtaceae), and found it effective for approximately 8 mo after treatment. The hemipteran pests can be expected to respond differently from *Josephiella* species. Although imidacloprid was not effective against stem gall wasps, it showed some effect against leaf gall wasps. We observed the effects of both imidacloprid and emamectin benzoate on leaf gall wasps for up to 18 mo after treatment, with emamectin benzoate being more effective than imidacloprid. The reduction in mean percentage of leaves infested on trees treated with emamectin benzoate was up to 31%. It is interesting that imidacloprid had an effect against leaf gall wasps but not against stem gall wasps.

Phosphorous acid is primarily used as a fungicide for the control of oomycetes. It is also considered to have some positive effect by stimulating the plant's natural defense system and overall health (Smillie et al. 1989). Therefore, we expected the phosphorous acid to have some effect against gall wasps through enhanced tree health and enhanced natural defense in combination with systemic insecticides, but the data showed no benefit of phosphorous acid in this particular tree–pest dynamic.

In conclusion, this study suggests that emamectin benzoate is effective for maintaining Chinese banyan tree canopy health for at least 14 mo under severe infestation with stem gall wasps, and for at least 18 mo under infestation with leaf gall wasps. Imidacloprid was only moderately effective against leaf gall wasps but not effective against stem gall wasps. Phosphorous acid did not provide any benefit to Chinese banyans in terms of suppressing these 2 wasp species. Overall, trunk injection with emamectin benzoate could be a feasible management strategy to control stem and leaf gall wasps on Chinese banyans in Hawaii, and possibly in other tropical regions of the continental U.S. where stem and/or leaf gall wasp species have established, such as Florida and California.

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