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Enhancing Ant-Termite Interactions to Protect Citrus Trees from Forest Tree Termite Damage

Introduction

Ants are an important component of most terrestrial ecosystems. Because of their high biomass, they interact with many other organisms. One such interaction is with termites, another organism with similar biomass that has considerable ecological effects. Among various types of interactions, ant predation on termites is the most important, widespread, and most studied type (Buczkowski and Bennett 2007). Understanding these interactions can have critical implications for minimizing the effects of pestiferous termites in forests, as well as in agro-ecosystems (Tuma et al. 2020).

Forest tree termites, *Neotermes connexus* (Blattodea: Kalotermitidae) have established colonies in Hawaiʻi (Grace 2010, Tong et al. 2017) and are normally found in forests, living in dead wood, wet wood, and live trees (Woodrow et al. 1999). The soldiers are large, around half an inch in length (Figure 1 top). In Hawaiʻi, this termite species is found to live in a wide range of fruit trees, especially causing damage to citrus (Sands 1977, Rouland-Lefèvre 2011). When the forest tree termite attacks fruit trees, they cause substantial economic losses for fruit growers.

The soldiers have prominent jaws that they use to excavate wood and make galleries inside young branches, where the colony subsequently resides and feeds on the wood. Older, infested branches are devoid of termites but break easily due to their generally weakened tree structure. Prolonged feeding by this termite species causes damage, resulting in limb breakage and tree collapse, leaving the decaying main stump on the ground (Figure 1 above, personal observations).

Upon close observations, we found this termite pest in close association with one common introduced ant species, the big-headed ant, *Pheidole megacephala* (Hymenoptera: Formicidae) (Figure 2 top). This ant species has heightened interspecific aggression, and is considered one of the "tramp" ant species (Passera 1994). Tramp ants relocate their nests frequently and can nest in various temporary nesting sites (Tay and Lee 2015). Because of these behaviors, they are very likely to become established in new habitats in both urban and agricultural settings. This ant occurs in large colonies that consist of major workers (with large heads) and minor workers (with small heads); the major workers have large mandibles for feeding (Warner and Scheffrahn, 2016).

Although some ant species are specialist predators, most of them are generalists and opportunist predators of termites (Tuma et al., 2020). Because termites present a rich source of lipids,

Figure 1. (top) Forest tree termites. (above) Damage caused by Forest tree termite feeding for multiple years, causing limb breakage and tree collapse.

December 2023
Subject Category: Insect Pests, IP-58

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This information has been reviewed by CTAHR faculty

proteins, minerals, sucrose, and micronutrients (Igwe et al 2011) – and the worker caste has soft bodies – they constitute suitable food for a wide range of animals, including ants (Figueirêdo et al. 2015).

In this study, we provided big-headed ants with a 25% sucrose solution, delivered by biodegradable alginate hydrogel beads, a naturally occurring polysaccharide derived from seaweed (Figure 2 above). The hydrogels were in bead-shaped form, and have the ability to absorb and hold any liquid. Once the ants find the hydrogels, the ants ingest the liquid directly from the hydrogel beads (Tay et al., 2017, 2020, Choe et al. 2021). The ants will then share the liquid with the entire colony. When these alginate beads are incorporated with 25% sucrose, they can provide an additional nutrient for the ants in the system. The sucrose hydrogel beads can biodegrade into the soil without any sticky, sugary residues in the environment (Schall et al. 2018, Tay 2023, Tay et al. 2023). When ants intake increasing amounts of sugar, they seek additional nitrogen to maintain a balanced protein-carbohydrate diet (Schumacher and Platner, 2009). Thus, we intended to examine whether the application of hydrogel beads laced with only sucrose solution would exploit the predatory potential of big-headed ants. It is hypothesized that the increased foraging behavior of big-headed ants would suppress the forest tree termite populations, thereby protecting citrus trees from termite damage.

Figure 2. (top) foraging big-headed ants searching for food and prey. (above) Hydrogel beads applied on the ground around the trunk of the citrus tree.

Methods

This study was conducted in a diversified fruit farm in Kilauea, Kaua'i from January to April 2021. On this farm, a citrus block consisting of around 30 Satsuma mandarin orange (*Citrus reticulata*) trees was chosen as a study site. Six trees each were randomly assigned to either the treatment (application of hydrogel beads containing 25% sucrose) or control (without application of hydrogel beads). Prior to the treatment application, we evaluated the tree status for the presence or absence of big-headed ants and the extent of forest tree termite damage. The suspected branches with termite damage were cut open using a hand saw to check for the presence or absence of termites. If the termites were observed, the damage was referred as the 'active' and if not, it was referred as the 'inactive' (Figure 3, top and above). Further, the main trunk of the tree was inspected for termite damage and the number of branches with termite damage was counted.

The hydrogel was produced in the laboratory according to the methods developed by Tay et al. (2017), but without any pesticides. Freshly manufactured and conditioned hydrogel beads were stored in the refrigerator until the day of treatment. The hydrogel beads containing 25% sucrose were applied to the treatment trees monthly for two consecutive months. For each application, approximately 50 grams of hydrogel beads were scattered on the ground around the base of orange trees near the trunk (Figure 2, above, Tay et al. 2023). Ant foraging was measured on each tree with a visual estimate of ant activity, in which the number

Figure 3. The damaged branches (top) Active, with the presence of termites inside. (above) Inactive, without termites.

of worker ants traversing a specific location on one of the side branches over a minute was counted. The ant counts were taken prior to the treatment, then 1, 5, 10, 23, and 31 days after treatment (DAT). The second application was made on the 31st day and the counts were taken 32, 42, 52, and 62 DAT. The hydrogel beads were applied on February 1 and March 4, 2023. The tree health status was assessed by examining the main trunk and the number branches for termite damage at the end of this study. A Termatrac® (termatrac.com) device was also used to detect whether active termites were inside the trunk. This device was placed with its radar surface against the trunk to record any signals produced by the movement of termites. In each tree, 2 to 5 recordings were taken around the trunk.

Results and Discussion

The damage caused by the forest tree termites in the orchard was visible before and after the study period. As in the natural environment, each of the study trees had a different health status and varying levels of termite damage, as indicated by the number of damaged branches (Table 1).

Prior to treatment, the mean number of big-headed ants was similar on treatment and control trees (hydrogel: 38.50 ± 12.8, control: 30.50 ± 5.5; Table 2). The number of ants gradually increased on the trees with hydrogel application and was highest at mid-month, then declined to levels similar to those of the control after a month (Figure 4). The percentage increase in ant numbers was greatest in the trees with hydrogel application compared to the control trees on 10 and 42 DAT (Table 2). Overall, this study showed that ant numbers increased in the trees with hydrogel application when compared to the control. For the control group, which demonstrated natural seasonal ant activity, ant foraging activity decreased over time (Figure 4). Tree health status in terms of termite damage remained similar for both pre-treatment and at 62 DAT.

None of the control trees, but 3 out of 6 treated trees, were recorded with at least one positive signal using Termatrac®. The results were not consistent when repeated several times around the trunk of same tree. Despite it being a non-destructive termite detection method, this inconsistent result suggests that the Termatrac® device may not be a reliable tool for detecting termites that are suspected to nest deep in the trees and in windy or wet weather conditions.

The hydrogel beads provide additional sucrose supplement to the ants, which might have resulted in better colony health and increased numbers of foraging workers able to search for prey. The increased ant foraging activities were maintained for a month. This indicated that monthly application would be required to replenish the sucrose hydrogels depleted by the ants in order to maintain higher than usual ant foraging activities throughout the season. During the study period, we cut open the termite-infested branches, where we observed big-headed ants that had entered the galleries and appeared to be fighting with termites. Although we did not observe ants feeding on termites, they were maneuvering termites and displacing them from the area. These observations suggest that big-headed ants can be opportunistic predators of termites.

Table 1: The pre-treatment health status of Satsuma mandarin orange trees, in terms of presence of ants and termite damage.

Treatment	Tree	Presence of Big-headed ants	Termite infestation status	No. of branches infested with termites
Control	1	Yes	Inactive	2
	2	Yes	Active	3
	3	Yes	Active	4
	4	Yes	Inactive	4
	5	Yes	Inactive	>5
	6	Yes	Inactive	>5
Hydrogel sugar application	1	Yes	Active	3
	2	Yes	Inactive	5
	3	Yes	Active	4
	4	Yes	Active	3
	5	Yes	Inactive	>5
	6	Yes	Inactive	>5

In this small-scale experiment, the fruit grower was aware that big-headed ants are an introduced species and are themselves considered to be a pest. Increased foraging activity of big-headed ants can have negative impacts on natural biocontrol agents because ants are known to tend and protect hemipteran pests (aphids, scales, mealybug, psyllids etc.), thereby increasing the pest abundance and probability of causing related plant diseases. However, problems related with hemipteran pests were not an issue in this farm and the grower preferred a non-chemical method that may help reduce termite impacts in citrus.

Table 2: Mean numbers of ants (± SE) pre-treatment and percentage increase in ant numbers in Satsuma mandarin orange trees on 10 and 42 days after the first treatment (DAT).

Treatments	Pre-treatment	% Increase After 10 DAT	% Increase After 42 DAT
Control	38.50 ± 12.8	1.8	37.7
Hydrogel sugar application	30.05 ± 5.5	90.0	136.2

In future studies, manipulative field or lab studies can help find out the predatory potential of big-headed ants for controlling forest tree termites. It was challenging to detect any change in tree health after only two months of treatment. Thus, a long-term experiment and more direct observations will help determine whether there are any predatory interactions between big-headed ants and forest tree termites that can lead to the suppression of termite populations in the citrus orchards.

Acknowledgements

This study was supported by Kaua'i County Extension POW 23-071 on Invasive Pest Management (to Manandhar), and Department of Plant and Environmental Protection Sciences POW 16-940 (to Tay). We thank Dave Whatmore, a fruit farmer in Kilauea, who drew our attention of forest tree termite problems in his citrus orchard and allow us to conduct this research.

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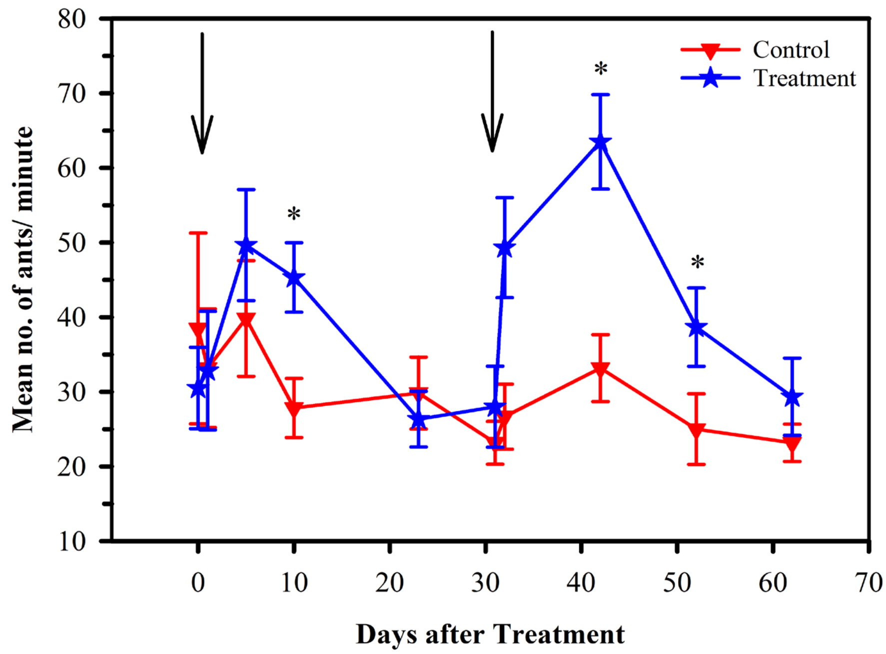

Figure 4. Mean number of big-headed ants in the Satsuma mandarin orange trees with hydrogel application and control treatments. Figure shows significantly different trends of big-headed ants foraging activity in the trees with hydrogel applications compared to controls (based on Proc Mixed, repeated measures analysis, SAS Institute), and significantly greater ant numbers was recorded on 10, 42, and 52 DAT (Proc Mixed, by date analysis, SAS Institute), as marked by asterisks. Arrows indicate the timing of hydrogel beads application.

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