APPENDIX 7-2: FINAL REPORT: SURVEY OF INVASIVE ANT SPECIES WITHIN MAKUA AND OAHU IMPLEMENTATION PLAN MANAGEMENT UNITS, OAHU, HAWAII 2004 - 2009

Final Report

Survey of invasive ant species within Makua and Oahu Implementation plan management units, Oahu, Hawaii 2004 - 2009

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Summary

Invasive ants have had devastating effects on biodiversity in the Hawaiian Islands, however information on new ant infestations and spread of existing populations into native communities is incomplete. Here we used bait cards to survey and identify invasive ant species on Makua and Oahu Implementation Plan Management Units on the island of Oahu. Twenty species of invasive ants were identified at 45 sites during 67 surveys conducted between 8 February 2004 and 7 October 2009. Survey sites ranged from sea level to 1220 m (4002 ft). Ants were abundant at most survey locations with the exception in some high elevation sites. Notably, the Mt Kaala boardwalk was surveyed twice and no ants were found on either occasion. As a result of nonrandom survey locations biased for upland habitat, the Papuan thief ant (*Solenopsis papuana*) was detected at more locations than any other species. The big-headed ant (*Pheidole megacephala*), yellow crazy ant (*Anoplolepis gracilipes*), and glaber ant (*Ochetellus glaber*) were also widespread. *Pheidole megacephala*, which tended to occur in the greatest numbers in areas it inhabited, compared to the other common species, and *Anoplolepis gracilipes* pose the most serious threats to intact native forests. Isolated infestations of these two species were identified and

control or eradication efforts may be warranted (see recommendations). Seventeen ant sampling locations containing five ant species (*P. megacephala*, *Plagiolepis alluaudi*, *O. glaber*, *S. papuana*, *Technomyrmex albipes*) were within a 50-m radius of known tree snail (*Achatinella* sp.) locations. There was a high degree of overlap among tree snails and *S. papuana*, possibly indicating some level of tolerance. However, other species of invasive ants were rarely surveyed in proximity to tree snail populations. These sites need to be carefully monitored as they may represent new invasions (especially in the case of *P. megacephala* along the fenceline at Ohikilolo) that could be addressed using ant control methodology. Based on the catastrophic effects ants have on many other invertebrates paired with anecdotal observations of ants negatively affecting tree snails reported here, research on the potential impacts of ants on tree snails is warranted to effectively conserve tree snails on Oahu and elsewhere.

Introduction

Ants (Hymenoptera: Formicidae) are not represented in the native fauna of the Hawaiian Islands (Wilson 1996). Since humans began to colonize the archipelago over 45 species of ants have been introduced (Nishida 1992; Krushelnycky *et al.* 2005; http://www.antweb.org/Hawai`i.jsp) and some species have had devastating effects on native ecosystems (Perkins 1913; Zimmerman 1970; Howarth 1985; Cole *et al.* 1992; Gillespie and Reimer 1993; LaPolla *et al.* 2000; Krushelnycky and Gillespie 2008). Invasive ants can reduce, extirpate and possibly cause the extinction of arthropod species (Perkins 1913; Zimmerman 1948; Solem 1967; Risch and Carroll 1982; Cole *et al.* 1992, Gillespie and Reimer 1993; LaPolla *et al.* 2000; Hill *et al.* 2003; O'Dowd *et al.* 2003; Plentovich 2010), directly and indirectly harm vertebrates (Meek 2000; Holway *et al.* 2002; Davis *et al.* 2008; Davis *et al.* 2009; Matsui *et al.* 2009; Plentovich *et al*. 2009) and alter plant communities (Bach 1991; Green *et al.* 1997; Christian 2001; Hill *et al.* 2003; O'Dowd *et al.* 2003; Handler *et al.* 2007). In some instances invasive ants can alter the overall structure and functioning of natural communities (Christian 2001**;** Holway *et al*. 2002; Hill *et al.* 2003; O'Dowd *et al*. 2003; Krushelnycky and Gillespie 2008).

Due to the small size and cryptic nature of many ant species, information on species distribution in the Hawaiian Islands is limited (Wilson 1996). On the main islands, there seems to be lower abundances and fewer species at higher elevations, and some areas at the highest elevations remain uninvaded (Krushelnycky *et al.* 2005). Since the majority of Hawaii's native biota is confined to high elevation areas, there is a need to identify which ant species are present in native forests, and then identify threats those species pose to native biota. Once this information is available we can determine whether control or eradication efforts are warranted.

Although there is much evidence that invasive ant species [e.g., the yellow crazy ant (*Anoplolepis gracilipes*), the big headed ant (*Pheidole megacephala*), Argentine ant (*Linepithema humile*)] harm a variety of native invertebrates (Cole *et al.* 1992; Gillespie and Reimer 1993; LaPolla *et al.* 2000), there is no experimental information on the effects of ants on endangered tree snails (*Achatinella* sp.) which can be found within the Makua and Oahu Implementation plan management units. Solem (1976) observed that tree snails were not found in areas with "swarming ants", especially *P. megacephala*. Professor Robert Cowie and Dr. Ken Hayes have reported similar observations throughout the main Hawaiian Islands (pers. comm.). Solem (1976) believed ants would not be able to penetrate the apertural barrier of adult snails. Therefore, he attributed the absence of snails in areas invaded by ants to predation of juveniles. This seems logical, however there is also an observation of ants attacking adult snails. Professor Michael Hadfield observed an unknown ant species attacking adult tree snails (*Achatinella* sp.) that were being kept in a greenhouse in Lyon Arboretum in upper Manoa Valley, Oahu. These attacks caused biologists to move the snails to a different location where access by ants could be limited (M. Hadfield pers. comm.). These anecdotal observations indicate that at least some species of ants do attack tree snails and that they may be a factor in reducing populations. It is unclear whether all invasive ant species are potentially harmful to native snail populations.

The objectives of this effort were to 1) identify potentially harmful ant species present on Makua and Oahu Implementation plan management units, 2) provide locale data that can be used to determine

which species overlap with known populations of threatened and endangered species, and 3) discuss ways to monitor sites for new infestations and to limit the spread of harmful species.

Methods

Survey Methodology: Index cards (12.7cm X 7.6cm) baited with potted meat (i.e., SPAM®), peanut butter, and honey were set in 45 sites on U.S. Army lands throughout Oahu between 8 February 2004 and 7 October 2009 (Figure 1). Survey locations ranged in elevation from sea level to 1220 m (4002 ft). The number of bait cards used at survey locations ranged from one to 30, depending on time constraints. Surveys often involved setting bait cards along an elevation gradient. A global positioning system was used to record longitude, latitude and elevation at each bait card unless forest cover precluded satellite acquisition. Surveys focused on areas with known populations of threatened and endangered species. Within these sites, special emphasis was placed on locations potentially serving as points of entry for invasive ants such as helipads, fence lines, camps, and out-planting sites. Survey coordinates were imported into ArcInfo and combined with locations of existing endangered tree snail populations to assess extent of overlap of invasive ants with tree snails.

Data Analysis: Density of individuals of each of the 4 common invasive ant species was compared using a Kruskal Wallis test. A nonparametric test was chosen because data were not normally distributed. Analyses were performed using JMP version 8 (SAS Institute Inc.).

Results

Twenty species of invasive ants were identified at 45 sites during 67 surveys conducted between 8 February 2004 and 7 October 2009 (Table 1, see Appendix 1 for raw data). Sites ranged in elevation from 6 m (19.7 ft) to 1220 m (4002 ft). *Solenopsis papuana*, *P. megacephala*, *Ochetellus glaber*, *A. gracilipes* and *Plagiolepis alluaudi* were the most commonly surveyed species respectively (Table 1). *Solenopsis papuana*, also called the Papuan thief ant, occurred at more than twice as many sites as the other ant species, however sampling effort was concentrated in upper elevation sites where habitat is more

suitable for this species compared to other common species (Table 1). Nineteen surveys were done in low elevation sites from sea level to 406m (0 – 1,333 ft), five in mid elevation forests from 406 to 812 m (1,333 to 2,666 ft) and forty-three in upland forests above 812m (2,666 ft). A comparison of densities of each species within invaded areas showed that, although *P. megacephala* only occurred at 12 sites, it tended to occur in the greatest numbers in areas it inhabited compared to the other 3 common species (Kruskal-Wallis Test, ChiSquare = 9.9, df = 3, P = 0.018, Table 1).

Very few areas remained uninvaded by ants. Cards with no ants were found between 396.2m (1300ft) and 1219.2 meter (4000ft). The Mount Kaala boardwalk was the only site where ants were not found in repeated surveys. It is possible that other areas such Makaha (above 2200 feet) and the Helemano Fenceline, are ant free, but additional surveys are needed to verify this.

Seventeen ant sampling locations containing five ant species (*P. megacephala*, *P. alluaudi*, *O. glaber*, *S. papuana*, *Technomyrmex albipes*) were within a 50-m radius of known snail locations. *Solenopsis papuana* occurred at nine of the 17 locations, *P. megacephala* occurred at three, *P. alluaudi* at 2 and both *T. albipes* and *O. glaber* at one, and no ants were found at one.

Discussion

Twenty species of ants were found from sea level to 1112.8m (3,651 ft). Despite the general conception that some species of ants are limited to lowland areas, we found four of the five most common species (*P. megacephala*, *O. glaber*, *A. gracilipes* and *P. alluaudi*) distributed throughout the sampling area. *Solenopsis papuana* was the most commonly sampled species, but was confined to mid and high elevation sites. Unlike *S. papuana*, in elevations above 880m (2890 ft), *A. gracilipes* and *P. megacephala* appear to be confined to isolated sites disturbed by humans. *Anoplolepis gracilipes* was first sampled in January 2008 at the Nike Greenhouse in the Waianae Mountains. Multiple site visits suggest that the *A. gracilipes* infestation is confined to a relatively small (<1 acre) area within and around the greenhouse. *Pheidole megacephala* was found on at least three occasions in 2008 at Ohikilolo above 880 m (2890 ft).

This infestation may be confined to area around the fence-line, but additional surveys are needed to identify the boundaries of the infestation.

The presence of *A. gracilipes* and *P. megacephala* at high elevations in or near some of the last intact native forest is troubling. Although we do not have experimental evidence, observations by Solem (1976) and Hadfield (per comm.) indicate that some invasive ant species might cause declines in tree snails via depredation of adults, eggs, and juveniles. Solem (1976) believed ants would not be able to penetrate the apertural barrier of adult snails and therefore ants. Therefore, he attributed the absence of snails in areas invaded by ants to predation of juveniles. Additional observations by Professor Michael Hadfield confirmed that ants also attack adult tree snails.

There is significant overlap between endangered snail populations and *S. papuana*. Gillespie and Reimer (1992) observed extensive overlap between *S. papuana* and native spiders (*Tetragnatha* sp). They found a significant inverse relationship between the abundance of S. *papuana* in an area and the diversity of native spiders. In the present study, the quality of data collected on ants and tree snails precludes a similar analysis, however, it is possible that, although *S. papuana* does coexist with tree snails, the species may still have some negative effects. Regardless, there is currently no feasible way to eradicate *S. papuana* at this time.

The extirpation of native invertebrates by invading ants tends to occur soon after invasion. A study in Australia showed that the longer *P. megacephala* was present in study sites, the lower the macro-invertebrate abundance (Hoffmann and Parr 2008). Krushelnycky and Gillespie (2008) show that endemic species are lost soon after invasion by alien ant species. Species that survive the initial invasion tend to be relatively resistant to the invader. *Solenopsis papuana* was first recorded in Hawaii in 1967 (Krushelnycky *et al.* 2005). It is possible that tree snails and other extant invertebrates are somewhat resistant to extirpation by *S. papuana*. Although negative effects may sill occur, as observed by Gillespie and Reimer (1992), extant tree snails may be less likely to be extirpated by *S. papuana* since they survived the initial invasion.

Preventing new ant invasions into relatively intact habitat in Hawaii and specifically, within the Makua and Oahu Implementation plan management units, is vital for the future of those native communities. This can be accomplished with careful monitoring of sensitive sites and adjacent areas where introductions are likely to occur. Sites requiring special attention may include, but are not limited to camping areas, trails, fence lines, helipads, and roads. Many harmful invasive ant species, such as *P. megacephala* and *A. gracilipes* primarily reproduce via budding (i.e., mated females walk rather than fly to nearby areas to found colonies) vs. mated flights. In these cases it is relatively easy to identify areas of encroachment by invasive ants into native forest.

Recommendations

1) Map the boundaries of the *A. gracilipes* infestation at the Nike Site. This can be accomplished by either setting a grid of bait cards or, if ant numbers are high enough, by having 3 people walk the boundary of the infestation; the inside person staying within the infestation, the outside person staying outside the infestation and the middle person recording waypoints along the boundary. This method has been used on Christmas Island, Indian Ocean and in Australia to successful map invasion boundaries.

2) Attempt to eradicate *A. gracilipes* from Nike Greenhouse site.

Begin by conducting a bait preference trail using a variety of ant baits with a sweet or fishmeal base. Put 1g of each product on small squares of paper/plastic side by side, and time how long it takes the ants to take the entire product. It may take half an hour for an “attractive matrix”. Another option is to weigh baits before and after leaving them sitting out for some period of time. The time period could range from 2 to 24 hours depending on the situation. Regardless of the method you choose, replicate at least 5 times. In general *A. gracilipes* is attracted to products with a fishmeal matrix, however, the species is known to be fickle, therefore, preference trials are necessary to maximize chances of successful eradication. Ideally the ants will be attracted to two different products, one with fipronil and the other with hydramethylnon as active ingredients. If this is the case, alternatively broadcast the products within the invaded area at 1 to 3

month intervals. Do three broadcasts starting and ending with the fipronil-based bait product. On Christmas Island, they found that low concentrations of fipronil (0.00015%) work as well as high concentrations (eg., 0.1%). You can expect about a 99% control rate for each broadcast. Spot treatments will be necessary to achieve eradication. It may also be possible to use bait stations instead of broadcast to eradicate this species.

3) Identify areas of encroachment by *P. megacephala* into native forest. Control using hydramethylnon suspended in a corn-grit matrix (eg., AMDRO®) if warranted. Apply according to label specifications.

4) Use bait cards to conduct yearly monitoring of sensitive areas so that any new infestations can be identified and addressed.

Ants are most likely to become established around disturbed areas frequented by humans such as bathrooms, campgrounds, fence lines, helipads, and roads. Areas undergoing construction of fences or other structures should be carefully monitored for new introductions. Activities including the transfer of soil, such as out-planting, should also be carefully monitored. Careful monitoring will increase chances of early detection, and early detection is the key to successful eradication or control.

5) Conduct additional surveys of high elevation sites in the Koolau Mountains.

High elevation sites in the Koolau Mountains were not well surveyed during this effort. Additional surveys are needed to identify ant species present in these areas and potential risks to resources.

6) Protect the Mount Kaala boardwalk area from invasion by ants.

Our data indicate that invasive ants have penetrated almost all areas with the exception of the highest elevation sites with intact native communities, such as the boardwalk area of Mount Kaala. Although ants were found at the gated entryway to the bog, none were found along the boardwalk. Every effort should be made to keep ants from penetrating this habitat. These efforts should include limitation of any activities, such as fence building that include disturbance. If these activities must take place, careful

monitoring of invasive ants should occur and may be necessary to control some species until disturbed habitat is allowed to recover.

7) Conduct additional research on the effects of invasive ants on tree snails.

Based on the catastrophic effects ants have on many other invertebrates, research on the potential impacts of ants on tree snails is necessary to effectively conserve these species on Oahu and elsewhere. Comparison of tree snail numbers in sites where ant densities are experimentally reduced vs. unmanipulated sites could reveal negative effects of ants on tree snails. Ant species should be carefully considered, as some ant species such as *A. gracilipes* and *P. megacephala* are likely to be far more detrimental than more cryptic species that occur at lower densities such as *Cardiocondyla venustula* and *C. emeryi*.

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Table 1. Alphabetical listing of twenty species of invasive ants, number of locations surveyed, average density (± mean standard error), and elevation range of detections within Makua and Oahu Implementation plan management units, Oahu, Hawaii from 2004 to 2009.

Genus	Species	# Locations	Average Density	Elevation Range (m)	Elevation Range (ft)
Anoplolepis	gracilipes	10	18.6±16.7	18.6 – 664.9	61-2180
Camponotus	variegatus	3	3	36.5 -664.9	120 - 2180
Cardiocondyla	minutior	1	1	1064.1	3489
Cardiocondyla	venustula	6	1.17±0.17	610 – 1112.8	2000 - 3651
Cardiocondyla	wroughtoni	4	1	610 - 1063	2000 - 3489
Cardioconyla	emeryi	1	2	686.2	2250
Leptogenys	falcigera	5	NA	6.1 – 634.4	20 - 2080
Monomorium	floricola	1	21	264.1	866
Monomorium	pharaonis	1	NA	854	2080
Ochetellus	glaber	15	126.8±52.37	12.2 – 902.8	40 - 2960
Paratrechina	bourbonica	1	NA	166.8	547
Paratrechina	longicornis	4	11.0±10.0	15.2 – 555.1	50 - 1820
Paratrechina	vaga	1	12	819.9	2690

Pheidole	megacephala	12	374±107.5*	264.1 – 664.9	866-2950
Plagiolepis	alluaudi	9	17.5±5.4	80.5 – 854	264 - 2800
Solenopsis	geminata	2	NA	594.7 -610	1950 -2000
Solenopsis	papuana	31	107.2±33.0	390.4 – 1018.7	1280 - 3340
Tapinoma	melanocephala	1	1	664.9	2180
Technomyrmex	albipes	7	192.5±126.2	9.1 -677	30 - 2220
Tetramorium	simillimum	6	17.0±7.7	264.1- 899.7	866 - 2950

* = species occurs at significantly higher densities than other species (Kruskal-Wallis Test, ChiSquare = 9.9, df = 3, P = 0.018)

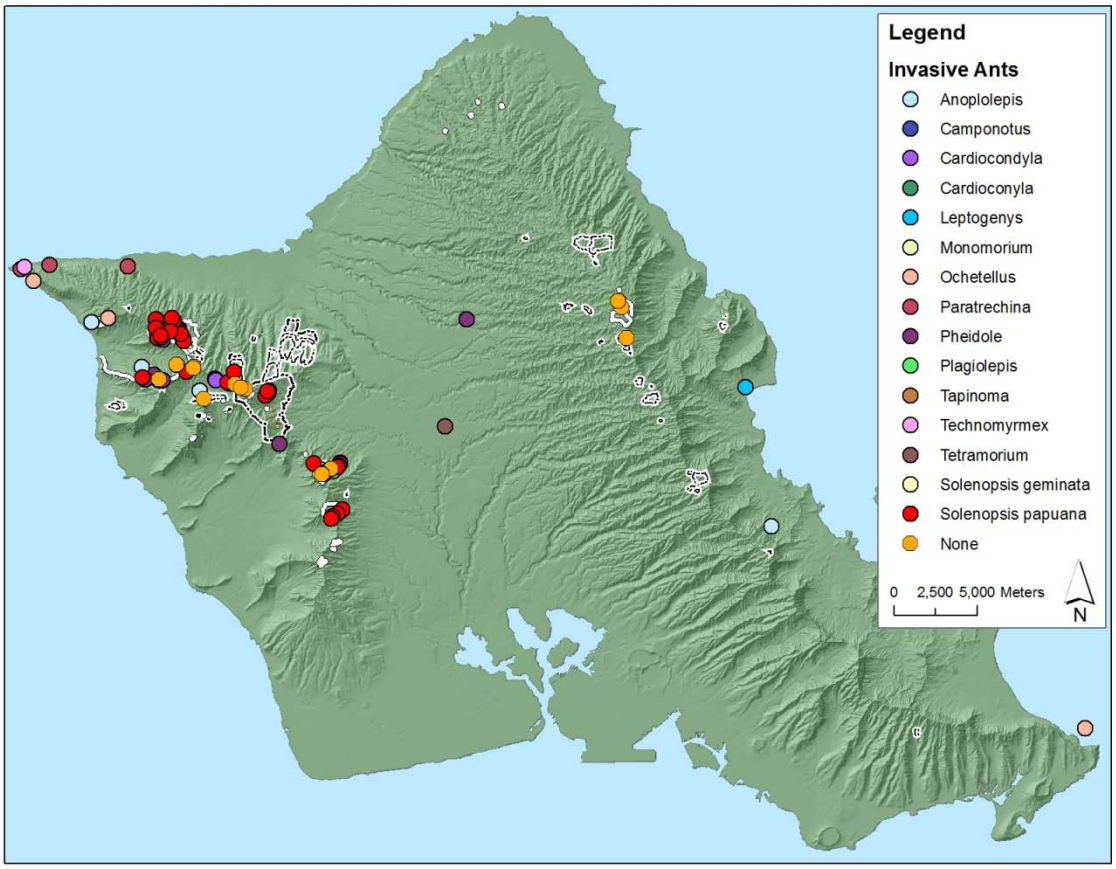

Figure 1. Distribution of ant surveys by species on the island of Oahu.

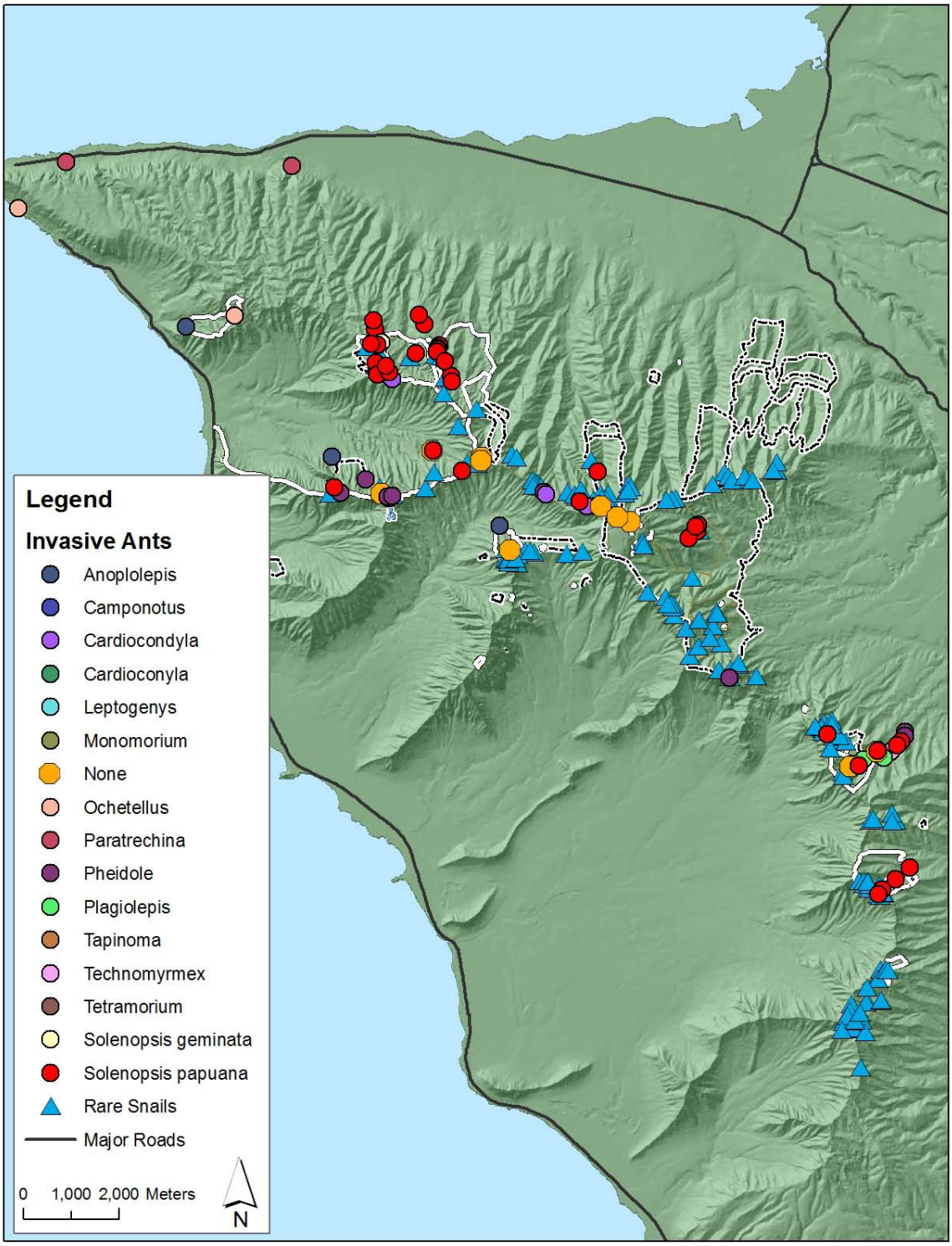

Figure 2. Location of invasive ant detections and proximity to known tree snail (*Achatinella* sp.) snail populations in the Waianae Mountains, Oahu.

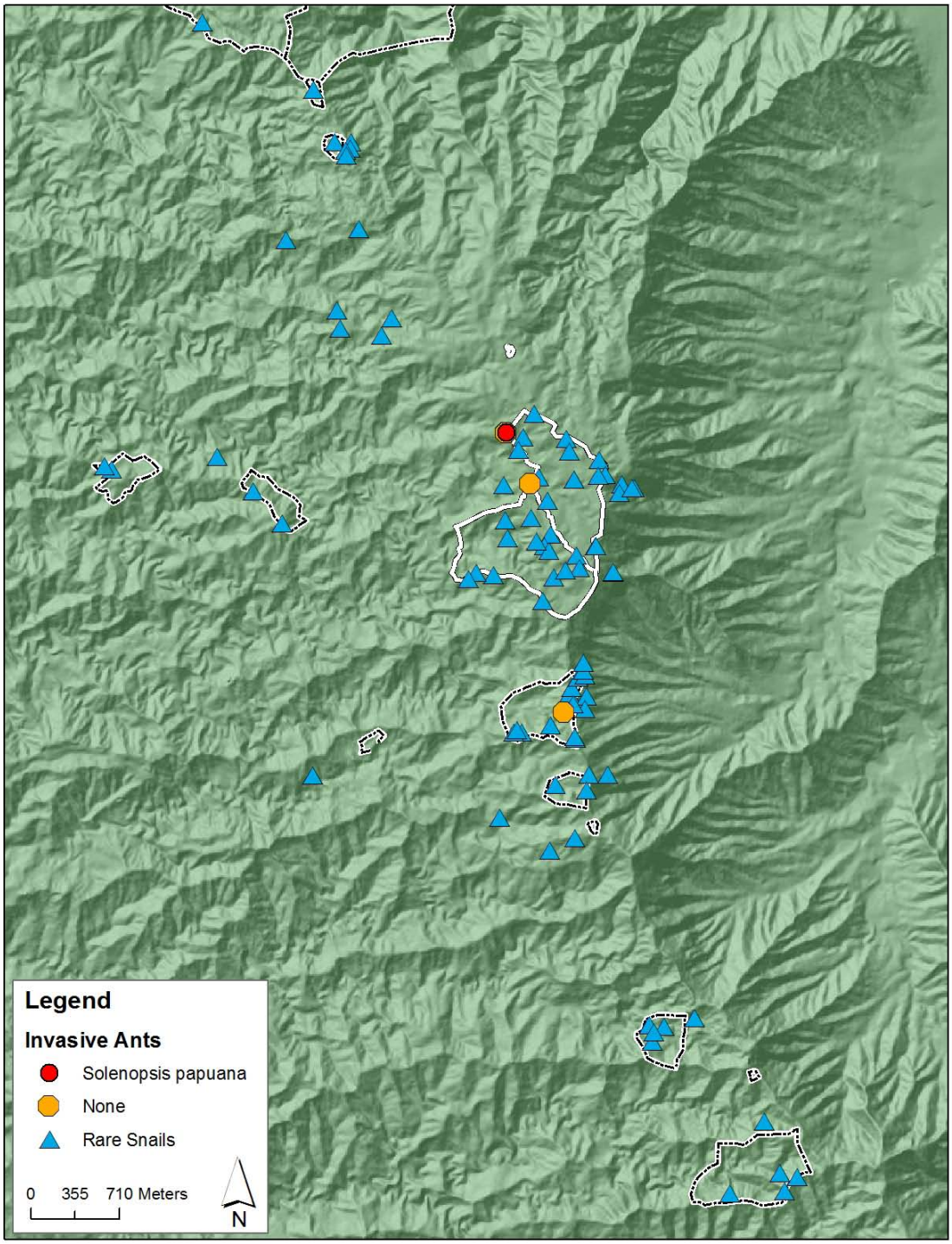

Figure 3. Location of invasive ant detections and proximity to known tree snail (*Achatinella* sp.) populations in the Koolau Mountains, Oahu.