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HONEYDEW-PRODUCING HEMIPTERANS IN FLORIDA ASSOCIATED WITH *NYLANDERIA FULVA* (HYMENOPTERA: FORMICIDAE), AN INVASIVE CRAZY ANT

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Abstract

Nylanderia fulva (Mayr) (Formicidae) is an invasive pest ant that has been reported in Florida, Texas, Louisiana and Mississippi. Workers tend various honeydew producing hemipterans in Florida landscapes and natural areas. We sought to understand the seasonal foraging activities of *N. fulva* and its relationship with honeydew producing hemipterans. Twenty-two hemipteran species were collected from 15 different plant hosts sampled in Florida from Jul 2010 to Jul 2012. The relative density of hemipterans on 4 plant species [live oak (*Quercus virginiana* Mill.; Fagales: Fagaceae), holly (*Ilex cornuta* Lindl.; Aquifoliales: Aquifoliaceae), magnolia (*Magnolia grandiflora* L.; Magnoliales: Magnoliaceae), and sugarberry (*Celtis laevigata* Willd.; Urticales: Ulmaceae)] was compared to the relative density of *N. fulva* at the base or stem of the selected plants. The number of *N. fulva* and hemipterans on each plant species was positively correlated from spring through fall. Both *N. fulva* and hemipteran populations increased from May to Sep and decreased from Oct to Apr. In addition, *Cinara juniperivora* (Wilson) (Hemiptera: Aphididae) on southern red cedar and *Toumeyella liriodendri* (Gmelin) (Hemiptera: Coccidae) on magnolia were covered with carton shelters presumably constructed by *N. fulva*, suggesting that this ant potentially protects certain hemipterans species.

Key Words: *Paratrechina pubens*, tawny crazy ant, Caribbean crazy ant, Rasberry crazy ant, Hemiptera, invasive ants, carton shelter

Resumen

Se ha reportado la presencia de *Nylanderia fulva* (Mayr), una hormiga plaga invasora en la Florida, Texas, Louisiana y Mississippi. Los trabajadores atienden varios hemípteros que producen rocio de miel en los campos y áreas naturales de la Florida. Tratamos a comprender las actividades estacionales de alimentación de *N. fulva* y su relación con los hemípteros que producen rocio de miel. Veinte y dos especies de hemípteros fueron recolectadas sobre 15 plantas hospederas diferentes muestreadas en Florida desde el julio de 2010 hasta el julio de 2012. Se comparó la densidad relativa de los hemípteros sobre 4 especies de plantas [roble vivo (*Quercus virginiana* Mill: Fagales: Fagaceae), el acebo (*Ilex cornuta* Lindl; Aquifoliales:. Aquifoliaceae), magnolia (*Magnolia grandiflora* L.; Magoliales: Magnoliaceae), y el almez de Mississippi (*Celtis laevigata* Willd; Urticales:. Ulmaceae)] con la densidad relativa de *N. fulva* en la base o tallo de las plantas seleccionadas. Se correlacionó positivamente el número de *N. fulva* y hemípteros en cada especie de planta desde la primavera hasta el otoño. La poblacion de *N. fulva* y los hemípteros incrementó desde mayo hasta septiembre y decrimentó de octubre a abril. Además, los *Cinara juniperivora* (Wilson) (Hemiptera: Aphididae) sobre el cedro rojo del sur y los *Toumeyella liriodendri* (Gmelin) (Homoptera: Coccidae) sobre la magnolia fueron cubiertos con refugios de cartón supuestamente construidos por *N. fulva*, lo que sugiere que esta hormiga potencialmente protege ciertas especies de hemípteros. Este es el primer informe de la construcción de refugios por *N. fulva*.

Palabras Clave: *Paratrechina pubens*, hormiga loca del Caribe, hormiga loca de frambuesa, Hemiptera, hormigas invasoras, refugio de cartón

Nylanderia fulva (Mayr) (Formicidae) is an invasive pest ant from South America that heavily infests buildings and landscapes in Florida, Louisiana, Mississippi and Texas (MacGown & Layton 2010; Warner & Scheffrahn 2010; Aguillard et al. 2011). The ants in Florida were originally identi-

TABLE 1. HEMIPTERANS TENDED BY *NYLANDERIA FULVA* IN GAINESVILLE, FLORIDA, FROM JUL 2010 TO OCT 2010.

	Insect Species		
Family	Scientific Name	Common name	Host plant
Aleyrodidae	*Dialeurodes citri* (Ashmead)*	Citrus whitefly	Ligustrum
Aphididae	*Aphis craccivora* Koch*	Cowpea aphid	Hemp sesbania
	Aphis vernoniae Thomas		
	Chaitophorus vimincola Hille Ris Lambers	Willow aphid	Black willow
	Cinara juniperivora (Wilson)	Juniper aphid	Southern red cedar
	Sanbornia juniperi Pergande		Southern red cedar
	Shivaphis celti Das*	Asian wooly hackberry aphid	Sugarberry
Coccidae	*Ceroplastes floridensis* Comstock*	Florida wax scale	Burford holly
	Neopulvinaria innumerabilis (Rathvon)	Cottony maple scale	Virginia creeper
Kermesidae	*Allokermes* sp.	Kermes scale	Live oak
Pseudococcidae	*Antonina graminis* (Maskell)*	Rhodes grass mealybug	St. Augustine grass
	Antonina pretiosa Ferris*	Noxious bamboo mealybug	Switch cane
	Dysmicoccus brevipes (Cockerell)*	Pineapple mealybug	Live oak
	Phenacoccus parvus Morrison*	Lantana mealybug	Live oak
	Palmicultor lumpurensis (Takahashi)*	Bamboo mealybug	Switch cane
Psyllidae	*Pachypsylla venusta* (Osten-Sacken)	Hackberry petiole gall psyllid	Sugarberry
Tingidae	*Corythucha floridana* Heidemann	Florida oak lace bug	Live oak

*Indicates introduced hemipteran insects to U.S.

Fig. 1. Intact and broken carton shelters showing tuliptree scale (*Toumeyella liriodendri* (Gmelin)) in magnolia (*Magnolia grandiflora* L.).

fied as *N. pubens* (Forel) and have been called the brown crazy ant, the Caribbean crazy ant, and the hairy crazy ant (Wetterer & Keularts 2008; Warner & Scheffrahn 2010; Calibeo & Oi 2011). The ants from Texas and Mississippi were called the Rasberry crazy ant, and were designated as *Nylanderia* sp. near *pubens* (Meyers & Gold 2008; MacGown & Layton 2010). However, Zhao et al. (2012) determined that all the specimens of *Nylanderia* species in both Texas and Florida are identical and Gotzek et al. (2012) confirmed that the pestiferous ants identified as *N. pubens* populations were actually *N. fulva*. *Nylanderia fulva* has caused electrical short circuits in Texas (Meyers 2008), destroyed and occupied bee hives in Texas (Harmon 2009), bitten people, and has displaced some native ant species in Colombia and Texas (Meyers 2008). *Nylanderia fulva* also has caused problems in parts of South America (Zenner-Polania 1990) and the Caribbean Islands because of their abundance and mutualism with hemipteran populations that damage crops, such as coconut (Wetterer & Keularts 2008).

Mutualisms between ants and honeydew producing hemipterans (e.g., aphids, scales, mealybugs, leafhoppers) are well-documented. Ants benefit from their association with hemipterans because their honeydew is an important source of carbohydrates (Stout 1979; Anderson & McShea 2001; Moya-Raygoza & Larsen 2008; Vanek & Potter 2010). Furthermore, hemipterans can serve as a protein resource if they are directly consumed by ants (Rosengren & Sundström 1991; Sakata 1994; Gullan 1997).

Conversely, hemipterans that produce honeydew benefit when they are tended by ants. Ants protect hemipterans from natural enemies and they sometimes provide them shelter (Sheppard et al. 1979; Stout 1979; Anderson & McShea 2001; Moya-Raygoza & Larsen 2008; Vanek & Potter 2010), thus allowing more hemipterans to survive and reproduce (Fritz 1982; Bristow 1983; Fowler & MacGarvin 1985; Buckley 1987). In addition, hemipterans that live inside ant nests (e.g., root feeding aphids and soft scales) are protected from climatic extremes (Buckley

Fig. 2. Intact and broken carton shelters showing Juniper aphid (*Cinara juniperivora* (Wilson)) on Southern Red Cedar (*Juniperus silicicola* (Small)).

1987). Some ants have been reported to assume parental care of hemipteran nymphs, enabling adult female hemipterans to produce more offspring (Bristow 1983). The removal of honeydew by ants may improve the hemipteran's environment by reducing the accumulation of sooty mold that grows on honeydew (Fokkema et al. 1983; Haines & Haines 1978). Sooty mold can suffocate hemipteran eggs (Moya-Raygoza & Nault 2000), affect crawler settling (Bess 1958), and cause adult mortality (Way 1954; Das 1959). In this study, we identified hemipteran species that were associated with *N. fulva* and documented *N. fulva* foraging activity with seasonal hemipteran abundance.

Materials and Methods

Hemipterans Associated with *Nylanderia fulva*

From Jul to Oct 2010, colonies of *N. fulva* were observed in parks, natural areas and neighborhoods in Gainesville (Alachua Co.), Florida, for any tending behavior towards natural infestations of hemipterans on trees, shrubs and other plants. *Nylanderia fulva* trails on plants were visually followed until ants were seen tending individual hemipterans. Leaves or stems with the hemipterans were collected and insects and plants were identified by specialists at the Florida Department of Agriculture and Consumer Services, Division of Plant Industry (FDACS/DPI).

Seasonal Sampling

Nylanderia fulva populations and honeydew-producing hemipterans were monitored monthly from Feb 2011 to Jul 2012 at 3 sites in Gainesville, Florida. Four plant species [live oak (*Quercus virginiana* Mill.; Fagales: Fagaceae), holly (*Ilex cornuta* Lindl.; Aquifoliales: Aquifoliaceae), magnolia (*Magnolia grandiflora* L.; Magnoliales: Magnoliaceae) and sugarberry (*Celtis laevigata* Willd.; Urticales: Ulmaceae)] that were infested with at least 1 honeydew-producing hemipteran species, and had trails of *N. fulva* going up the stem were selected for monitoring. Four plants of each species were sampled except for magnolia, for which only one plant was sampled in 2011 and 3 more plants were added in 2012. The selected plant species were all located in managed landscapes, usually along driveways. Six to 10 terminal shoots per plant with at least 6 to 10 leaves were arbitrarily chosen, cut with a pole pruner and transported in plastic bags to the laboratory on ice. The total number of honeydew-producing insects on terminal shoots was counted within 24 h of collection.

Nylanderia fulva activity was estimated in 3 ways for each plant. First, a slice of sausage (0.5 × 2.5 cm Armour Original Vienna Sausage, Cherry Hill, New Jersey) was placed on the center of an index card (7.6 × 12.7 cm) that was positioned about 25 cm from the base of the plant on the northern and southern sides. After 15 min, each card with ants was individually bagged, frozen and the number of ants per sausage was determined. Second, the number of *N. fulva* foraging trails going up one major trunk per plant from the ground was counted. Finally, the number of workers walking past a specific point (~30.5 cm above the ground) on the most active foraging trail was counted for 20 s. All sampling occurred between 0900-1500 h when the air temperature was > 21 °C.

Pearson's correlation was used to examine the association between the total number of hemip-

TABLE 2. LIST OF TREES, SAMPLING LOCATIONS, INSECTS AND NUMBER OF LEAVES THAT WERE SAMPLED.

Tree	Site	Mean tree ht (m)	Honeydew producers	No. of leaves/ branch
Burford holly (*Ilex cornuta* Lindl.)	Site 1	0.76	Florida wax scale (*Ceroplastes floridensis* Comstock) Mealybug (*Dysmicoccus texensis* (Tinsley))	6
Live oak (*Quercus virginiana* Mill.)	Site 1	4.27	Lace bug (*Corythucha floridana* Heidemann) Aphids (*Myzocallis puncata* (Monell)) Kermes scale (*Allokermes* sp. (Cockerell)) Bullet gall (*Disholcaspis quercusvirens* (Ashm.)) Pineapple mealybug (*Dysmicoccus brevipes* (Cockerell))	10
Magnolia (*Magnolia grandiflora* L.)	Site 2 Site 3	2.29	Tuliptree scale (*Toumeyella liriodendri* (Gmelin))	3
Sugarberry (*Celtis laevigata* Willd.)	Site 2	6.4	Asian wooly hackberry aphid (*Shivaphis celti* Das) Flatid plant hopper Psyllid gall (*Pachypsylla* sp.)	10

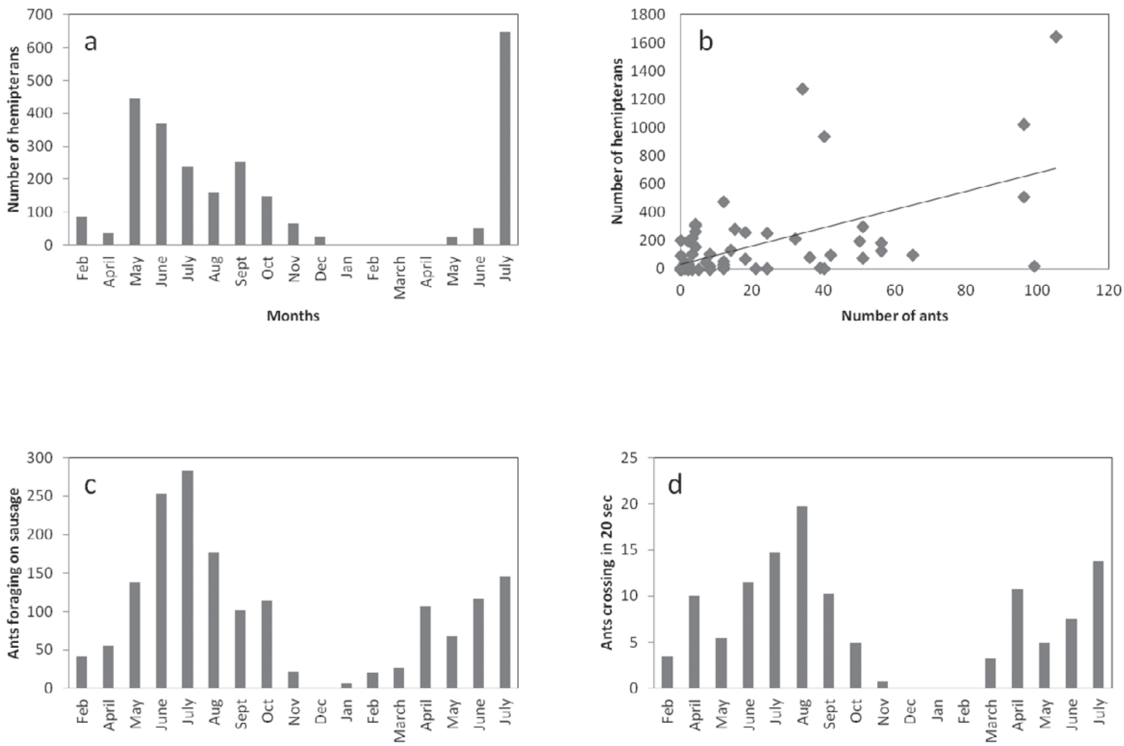

Fig. 3. a) Mean number of hemipterans collected from the shoots every month from holly in Gainesville from Feb 2011 to Jun 2012. b) Correlation between number of hemipterans and number of ants (trailing intensity) on holly (r = 57). c) Mean number of ants collected on sausage samples every month at the base of holly in Gainesville from Feb 2011 to Jun 2012. d) Mean trailing intensity (i.e. ants crossing a specific point in 20 s × number of trails) on holly from Feb 2011 to Jun 2012.

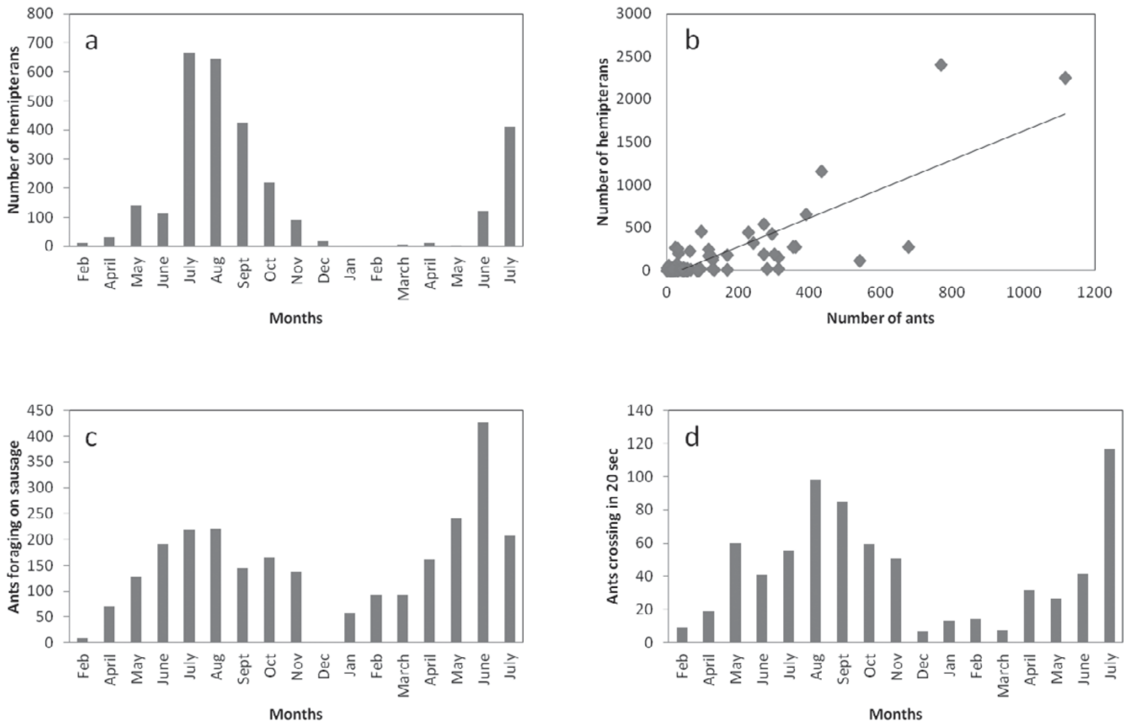

Fig. 4. a) Mean number of hemipterans collected from the shoots every month from live oak in Gainesville from Feb 2011 to Jun 2012. b) Correlation between number of hemipterans and number of ants (trailing intensity) on live oak (r = 0.80). c) Mean number of ants collected on sausage samples every month at the base of live oak in Gainesville from Feb 2011 to Jun 2012. d) Mean number of trailing intensity (i.e. ants crossing a specific point in 20 s × number of trails) on live oak from Feb 2011 to Jun 2012.

terans on a shoot and the 20 s ant count. Due to the increased number of trails per trunk in the summer, the 20 s count was multiplied by the number of trails to provide an index of trailing intensity. Analysis of variance (ANOVA) was conducted to determine differences in 20 s ant counts from the most active ant trail among plant species and Tukey's HSD test was used to conduct a mean separation among plant species (R Development Core Team 2012).

Results and Discussion

Hemipterans Associated with *Nylanderia fulva*

A total of 17 species from 7 families of honeydew-producing hemipterans were tended by *N. fulva* on 10 plant species (Table 1). We did not observe *Nylanderia fulva* exhibiting any obvious preference for any particular hemipteran species. Aphids and mealybugs were the most prevalent and had the greatest diversity of species, with 6 species of aphids and 5 species of mealybugs. Similar observations of greater species diversity for these 2 families have also been reported in previous studies. Zenner-Polania (1990) reported *N. fulva* tending whiteflies and scale insects in orange trees and mealybugs in rangeland grass, sugarcane and coffee berries. They also observed that the ants transported the hemipterans from infested to uninfested plants and protected the hemipterans from the predators by constructing protective shelters over the mealybugs.

We observed carton shelters on magnolia branches, where *N. fulva* had covered tuliptree scales using soil and plant debris (Fig. 1). Shelters were also observed on the trunk and near the base of the trunk where there were pruning injuries. *Nylanderia fulva* also built carton shelters around Juniper aphids (*Cinara juniperivora* (Wilson); Aphididae) along a split on the trunk on Southern red cedar (*Juniperous silicicola* (Small); Pinales: Cupressaceae) (Fig. 2). The structures were built in such a way that ants had access to enter and exit those shelters. When the carton was broken apart, many *N. fulva* workers scrambled out of the shelters and the juniper aphids or tuliptree scales (*Toumeylla liriodendri* (Gmelin); Coccidae) that were previously covered were vis-

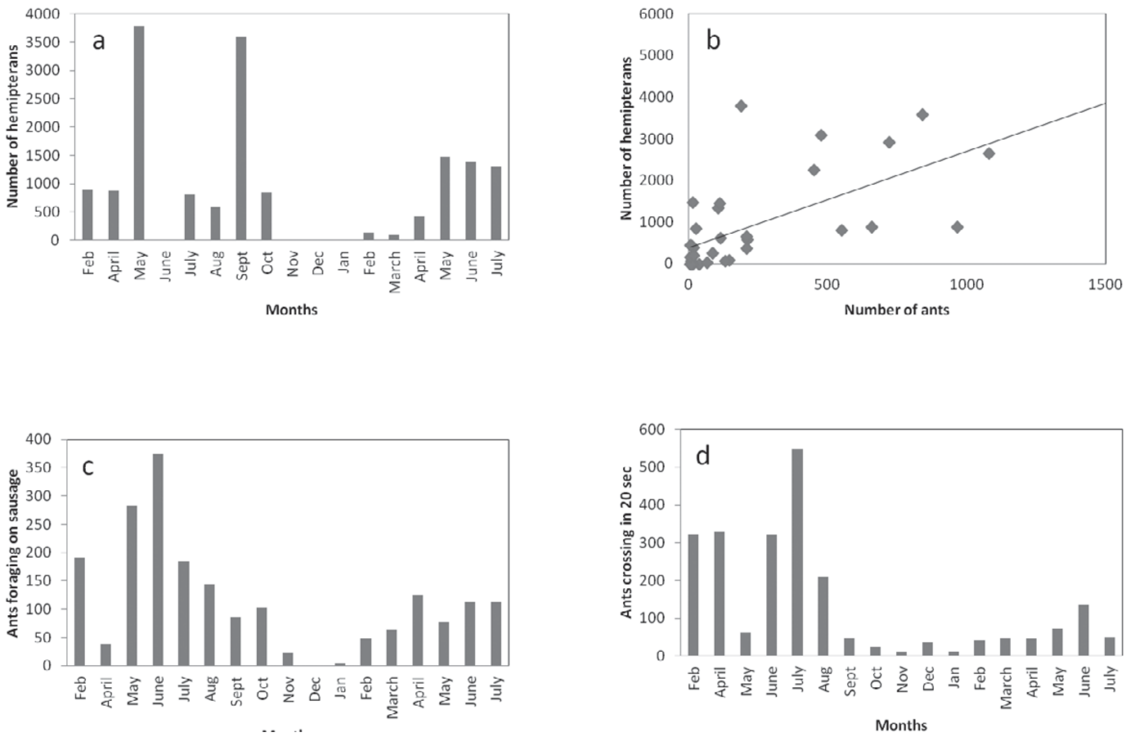

Fig. 5. a) Mean number of hemipterans collected from the shoots every month from magnolia in Gainesville from Feb 2011 to Jun 2012. b) Correlation between number of hemipterans and number of ants (trailing intensity) on magnolia (r = 0.63). c) Mean number of ants collected on sausage samples every month at the base of magnolia in Gainesville from Feb 2011 to Jun 2012. d) Mean number of trailing intensity (i.e. ants crossing a specific point in 20 s × number of trails) on magnolia from Feb 2011 to Jun 2012.

ible. Many ants, like *Acropyga*, *Formica*, *Lasius*, *Odontomachus* and *Oecophylla*, build protective shelters of soil and plant debris, or carton, over honeydew producing hemipterans (Wheeler 1910; Way 1963; Evans & Leston 1971). These shelters are thought to protect the hemipterans from adverse environments (Way 1963; Helms & Vinson 2002), thereby increasing hemipteran populations and the availability of honeydew for the ant colony (Dejean et al. 1997).

Seasonal Sampling

The hemipterans collected from plants that were sampled monthly from Feb 2011 to Jun 2012 are listed in Table 2. The predominant hemipteran species on each host plant were Florida wax scale, *Ceroplastes floridensis* Comstock (Coccidae), on holly; the aphid, *Myzocallis puncata* (Monell), on live oak; the tuliptree scale, *Toumeyella liriodendri* (Gmelin), on magnolia; and the Asian wooly hackberry aphid, *Shivaphis celti* Das, on sugarberry. *Nylanderia fulva* were observed tending 5 hemipteran species on live oak and 3 species on sugarberry. Two hemipterans were sampled from holly, and only one was on magnolia. Among the sampled tree species, live oak and sugarberry were large, mature trees while holly and magnolia were smaller and younger landscape plantings that were pruned (Table 2). Thus, the difference in plant age and architecture could have affected the diversity of herbivorous hemipterans.

Hemipteran populations were higher during the warmer months (May to Sep) and decreased as ambient temperatures dropped at the end of Oct (Figs. 3a-6a). Similar temporal patterns of hemipteran distribution have been reported by Barlow (1962) for *Myzus persicae* (Sulzer) (Aphididae) on potatoes, *Macrosiphum euphorbiae* (Thomas) (Aphidae) on tobacco, and by Dixon (1977) in walnut aphid, lime aphid, cereal aphids and black bean aphid. Stevens et al. (1998) found that the population of several ant species that were tending the honeydew producing hemipterans decreased in cooler months when hemipteran numbers declined and increased in warmer months when hemipterans returned.

Similarly, *N. fulva* activity was greater during the warmer months (e.g., May to Aug) on all 4 plant species (holly, love oak, magnolia and

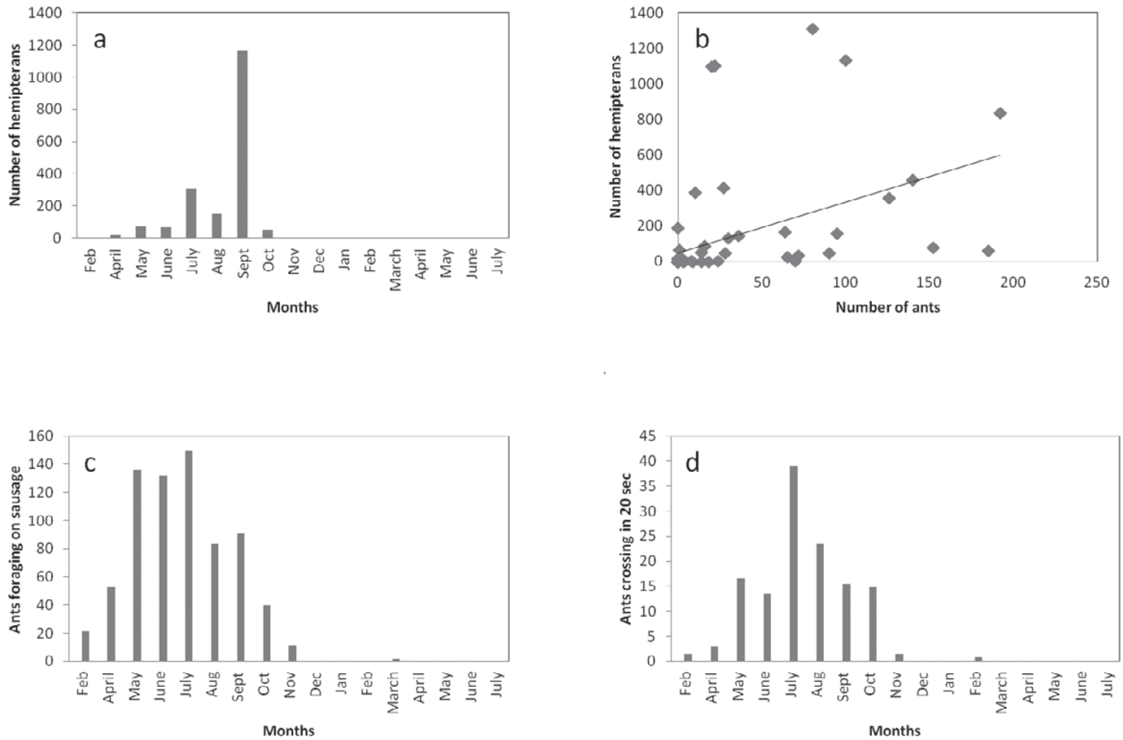

Fig. 6. a) Mean number of hemipterans collected from the shoots every month from sugarberry in Gainesville from Feb 2011 to Jun 2012. b) Correlation between number of hemipterans and number of ants (trailing intensity) on sugarberry (r = 0.44). c) Mean number of ants collected on sausage samples every month at the base of sugarberry in Gainesville from Feb 2011 to Jun 2012. d) Mean number of trailing intensity (i.e. ants crossing a specific point in 20 s × number of trails) on sugarberry from Feb 2011 to Jun 2012.

sugarberry). Figure 3c-6c shows the number of ants foraging on the sausage provided during the samplings. The average number of ants crossing a specific point in 20 s during the 16 mo period in 2011-2012 is presented in Figures 3d-6d. As the daily average temperature decreased in Oct and Nov (< 21 °C), ant numbers declined slightly. When the temperatures cooled further in Dec and Jan (< 19 °C), ant foraging decreased dramatically, suggesting that the *N. fulva* activity is largely dictated by temperature. Ants are poikilothermic, thus their foraging activity depends on different abiotic factors like relative humidity and soil temperature (Traniello 1989; Valenzuela-González et al. 1995). Porter & Tschinkel (1987) reported that *Solenopsis invicta* foraging activity was limited due to low temperatures in several locations of southeastern United States. *Nylanderia fulva*'s activity appeared to be less in 2012 than in 2011 during similar months (Figs. 3a,c,d-6a,c,d). This was possibly due to the lower average RH (< 30%) in 2012 than in 2011. In addition, the average precipitation for Dec to Apr was also lower in 2012 (3.6 cm) than in 2011 (7 cm). Hölldobler & Wilson (1990) suggested that warmer temperatures may not be enough for high ant activity. A relatively humid environment may be needed to avoid desiccation and to resume their activities in the open. Therefore, lower humidity and rainfall might have contributed to reduced *N. fulva* abundance in 2012 as compared to similar time periods in 2011. Hemipteran population size (Figs. 3a-6a) was also lower in 2012 than in 2011. It was not clear from our sampling alone that hemipteran numbers were lower because of decreased ant abundance or vice versa. More research will be required to evaluate this relationship.

Significant positive correlations were found between the total number of ants (trailing intensity) and the number of hemipterans in holly (r = 0.57, $P < 0.0001$), magnolia (r = 0.63, $P < 0.0001$), live oak (r = 0.80, $P < 0.0001$) and sugarberry (r = 0.44, $P < 0.0001$) (Figs. 3b-6b). Trees with higher hemipteran populations had higher *N. fulva* trailing intensity (i.e., the number of trailing ants crossing a specific point in 20 s × number of trails). Trailing intensity over time differed significantly among the 4 plant species (Fig. 7). Magnolia had an average trailing intensity of 136 ants throughout the sampling period which was significantly

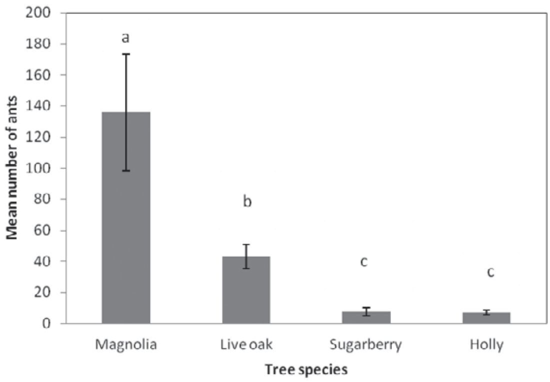

Fig. 7. Mean number of ants crossing a specific point in 20 s on different tree species shown by analysis of variance and mean separation by Tukey's HSD test. Standard error bars with different letters indicate significantly different values ($P < 0.05$) (R Development Core Team 2012).

higher than the 43 ants on live oak. The trailing intensity on sugarberry and holly were similar at 8 and 7 ants, respectively (Fig. 7). Greater *N. fulva* trailing on magnolia and live oak could be attributed to greater hemipteran diversity and abundance. Tuliptree scale in magnolia is present in all stages of development during the winter in southern states (Donley & Burns 1971) and even though most insect species found in live oak were present in warmer months, lace bugs were present throughout the year in areas with mild winters like Florida (Dreistadt & Perry 2006) providing a continuous supply of honeydew to *N. fulva*. In contrast, on holly and sugarberry hemipterans were found only during the warmer months (Jul to Oct), and as a deciduous tree, sugarberry was defoliated from Oct to Feb. Brightwell & Silverman (2011) determined that there were fewer *Linepithema humile* (Mayr) (Formicidae) nests around deciduous trees in fall, but were present around evergreen species *Pinus taeda* L. (Pinales: Pinaceae) throughout winter and successfully foraged in these trees even when ambient temperatures were below the minimum foraging threshold. This pattern of behavior may be similar for the deciduous trees in our study where *N. fulva* moved their nests away from sugarberry in the winter, thus reflecting the reduced number ants in sugarberry after Oct 2011 (Figs. 6a,c,d).

In this study, the activity periods of honeydew producing hemipterans and *N. fulva* were positively associated, which reflected the seasonal changes in temperatures. During the warmer months (May to Oct), the presence and abundance of hemipterans increased, which was associated with greater ant foraging. Our paper also documents that some species of honeydew producing hemipterans that were tended by *N. fulva* were apparently protected by shelters created by *N. fulva*. Thus, there is evidence of mutualism between *N. fulva* and honeydew-producing hemipterans. Understanding the seasonal phenology between honeydew-producing hemipteran species tended by *N. fulva* could be important in developing control strategies for this invasive ant. Controlling hemipteran species would remove a food resource of *N. fulva* and thus could help decrease the density of ants in the landscape.

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