Effect of Aromatic Cedar Mulch on Argentine Ant (Hymenoptera: Formicidae) Foraging Activity and Nest Establishment

HEIKE E. MEISSNER AND JULES SILVERMAN[1]

Department of Entomology, North Carolina State University, Raleigh NC 27695–7613

J. Econ. Entomol. 96(3): 850–855 (2003)

ABSTRACT In the laboratory, Argentine ant, *Linepithema humile* (Mayr), mortality was positively correlated to the length of an aromatic cedar mulch section that had to be crossed before food could be reached. When ants could access food without crossing the mulch, mortality was not correlated to mulch section length. In the field, Argentine ants showed a tendency to avoid aromatic cedar mulch as a nesting substrate. In plant beds alongside buildings the number of ant nests (pockets containing brood) found was not significantly different between aromatic cedar and cypress mulch. However, when pine straw mulch around oak trees was replaced with aromatic cedar or cypress mulch, a similar number of ant nests was found in the cypress mulch as in the original pine straw, whereas numbers in aromatic cedar mulch were significantly lower. Also, fewer ants were trailing on the trees surrounded by aromatic cedar mulch compared with cypress mulch or the original pine straw. The number of ants attracted to apple jelly baits placed alongside the buildings did not differ between mulch types; neither did the number of ant trails crossing the mulch beds around the buildings. We suggest that aromatic cedar mulch may help control Argentine ants and reduce insecticide input when applied in combination with conventional control methods.

KEY WORDS *Linepithema humile*, aromatic cedar mulch, cultural control, field study, nesting sites

THE ARGENTINE ANT, *Linepithema humile* (Mayr), a species of South American origin, has successfully invaded regions of the world with Mediterranean and mild temperate climates. In the United States, it is established in many southeastern states and in California (Newell and Barber 1913, Ward 1987), where it ranks among the species most frequently encountered in and around people's homes (Rust and Knight 1990, Vega and Rust 2000). Colonies consist of networks of numerous interconnected nests, frequently covering entire neighborhoods. Relatively fast population growth rates, diffuse nest structure and swift nest relocation following disturbance make Argentine ant infestations extremely difficult to control. Management efforts rely almost exclusively on insecticides, with limited success.

In addition to being a nuisance pest, the Argentine ant also presents a major ecological threat by displacing the native ant fauna of invaded areas (Ward 1987, Holway 1998, Kennedy 1998). For example, Christian (2001) demonstrated that Argentine ants are causing a decline of large-seeded plant species in the fynbos shrublands of South Africa by eradicating ants that act as seed dispersers for these plants. Numerous other studies also address the ecological consequences of Argentine ant invasions (Messina 1981, Beattie 1985, Bond and Slingsby 1984, Vega and Rust 2000).

Argentine ants may nest in a large variety of substrates (Passera 1994), but prefer the top 20 cm of well-drained, loose matter, moving the nest vertically as conditions change (Smith 1965, Markin 1968). In residential settings, they are frequently found in landscaping mulch (H.E.M., unpublished data). Mulch placement alongside buildings and around fruit trees is common and may contribute to the presence of ant colonies in locations where they are least desirable.

The wood of the eastern red cedar (also known as aromatic cedar), *Juniperus virginiana* L., contains volatile chemical constituents that appear to be toxic to some insects. In a previous study we showed that Argentine ants and odorous house ants, *Tapinoma sessile* (Say), avoided aromatic cedar mulch as a nesting substrate in the laboratory and died when confined to this mulch (Meissner and Silverman 2001). Other researchers have also shown lethal effects of aromatic cedar wood to the clothes moth, *Tineola bisselliella* (Hum.), (Scott et al. 1918, Laudani and Clark 1954), the black carpet beetle, *Attagenus piceus* (Oliv.), termites (Adams et al. 1988), and housedust mite, *Dermatophagoites spp.* (Enomoto et al. 1999). Repellency of the mulch was furthermore shown for the German cockroach, *Blattella germanica* (Linn.) (Appel and Mack 1989), and the red imported fire ant, *Solenopsis invicta* Buren (Thorvilson and Rudd 2001).

The effects of aromatic cedar mulch on Argentine ant populations in the field have never been tested. If

[1] E-mail: Jules_Silverman@ncsu.edu.

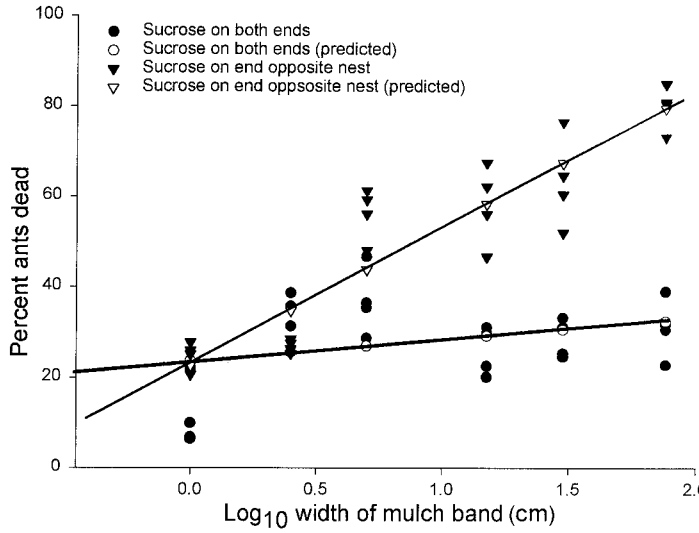

Fig. 1. Regression of percent ant mortality (5-d total) against log of aromatic cedar mulch section length for set A (sucrose on both ends of the arena) and set B (sucrose only on end opposite the nest). See Table 1 for regression statistics.

efficacious, this mulch may easily be incorporated into a residential ant management program to help reduce insecticide use in the proximity of human dwellings.

We assessed ant mortality caused by voluntary movement across aromatic cedar mulch in the laboratory. Furthermore, we determined the effectiveness of aromatic cedar mulch in suppressing Argentine ant foraging activity and preventing the establishment of nests outdoors.

Materials and Methods

Insects. Argentine ants used in this study were taken from laboratory colonies established from various field collections during 2000 in North Carolina. These colonies were kept in plastic trays (40 × 52 × 8 cm, Panel Controls, Greenville, SC), coated with Fluon (AD-1, Northern Products, Woonsocket, RI) to prevent escape. Ant colonies were provided with artificial nests, each of which consisted of a petri dish (15 mm diameter) filled with dental plaster which was sculptured to increase nest surface area, and glued on top of a water-filled glass jar, from which a cotton wick conducted water to the ants. A 25% sucrose solution, artificial diet (Nonacs and Dill 1988), and German cockroaches were provided as food ad libitum.

Mortality because of Movement Across Mulch. In laboratory experiments we determined whether Argentine ants would cross aromatic cedar mulch to access food, and we assessed the impact of contact with aromatic cedar mulch on survival. Plexiglas arenas (0.1 × 1.0 m) had a certain portion of their length (76, 30, 15, 5, 2.5, or 0 cm) covered with a 1 cm thick layer of aromatic cedar mulch (P&L Bark Inc., Pageland, SC). Regardless of their length, the mulch sections were centered within the arenas and covered their entire width.

Before adding mulch, 1,000 workers and two queens were placed inside one end of each arena with a nest. The nest was a glass test tube (10 cm × 2 cm diameter) about one-half filled with water and plugged with cotton, then wrapped with aluminum foil to exclude light. The ants were deprived of food for 24 h; then ants outside the nest were collected with an aspirator, mulch was placed, and the ants were released next to the nest. Ants were provided 25% sucrose (≈25 ml) in dispensers, positioned either one next to the nest and a second one across the mulch on the far end of the arena (set A), or only across the mulch (set B). Consequently, in set B ants had to cross the mulch to access the sucrose, while in set A they were able to feed without having to cross the mulch. Experiments were conducted at 27 ± 2°C, 50 ± 10% RH, and a photoperiod of 12:12 (L:D) h. Each treatment was replicated four times. Dead ants on the end opposite the release site were counted daily and removed from the arena. At the end of the experiment (day 5), the total number of dead and live ants in the arenas was counted and percentage mortality was calculated.

Effect of Mulch Placed Alongside Buildings. We determined in the field whether aromatic cedar mulch would reduce Argentine ant nest site selection, and thereby constitute an effective barrier to Argentine ant movement. We identified three large office buildings with a history of Argentine ant activity in an office park in Morrisville, NC. On 17–20 May 2001 we determined that the pine needle mulch in the plant beds surrounding these buildings was completely free of Argentine ant nests (defined as any accumulation of brood and workers), yet workers were generally trailing close by at all locations. We then replaced the pine needle mulch surrounding the buildings with a 5 cm thick layer of either aromatic cedar or cypress mulch (P&L Bark, Pageland, SC) in alternating lengths of

Table 1. Regression statistics for the effects of mulch section length on ant mortality

	df	slope (±SE)	t	P	R^2
Sucrose only on far end	1	303.9 (28.9)	10.50	<0.0001	0.83
Sucrose on both ends	1	46.4 (32.5)	1.43	0.17	0.04

10–30 m. Cypress mulch was selected for comparison, because it has a similar consistency to aromatic cedar mulch and was shown in previous laboratory experiments (Meissner and Silverman 2001) to be neither toxic nor repellent to Argentine ants. The total area covered with each mulch type was ≈200 m^2. The plant beds contained various ornamental shrubs, 0.5–1m in height, which did not touch the building walls. No insecticide was applied during the course of the study.

We counted the number of nests found in each mulch type on a biweekly basis between 27 July and 10 October 2001. Also, the number of ant trails (clearly visible lines of constant ant movement) crossing each mulch type was counted every other week. In addition, we placed baits (petri dishes containing apple jelly) alongside the buildings, either directly on the mulch or on a window sill (1 m high above the mulch). This was done once every other wk at a total of 39 locations per mulch type and bait height. The number of ants attracted to the baits was counted 2 h after bait placement, after which time the baits were removed.

Effect of Mulch Around Trees. In the same office park as above, 24 oak trees (3–5 m high, and 20m apart), planted in a row alongside a parking lot and each surrounded by a ring of pine needle mulch (1 m wide and 30 cm high), were assigned equally (eight replicates) to the following three treatments: 1) pine needle mulch replaced with cypress mulch; 2) pine needle mulch replaced with aromatic cedar mulch; 3) pine needle mulch left in place undisturbed (control). Presence or absence of Argentine ant nests in the pine straw surrounding the trees had been recorded before the start of the experiment (3 August, 2001), and each treatment was assigned five trees with and three trees without ant nests; however, all trees had ants trailing along the trunk from the base to the canopy. During the experiment (between 9 August and 5 October 2001) we conducted biweekly inspections for presence of ant nests in the mulch. Furthermore, we carried out biweekly counts of the number of ants per min crossing (in both directions) an imaginary line surrounding the tree trunk at a height of 1 m. No insecticide was applied during the course of the experiment, and the mulch was left undisturbed by us, except during data collection.

Statistical Analysis. For each set of the laboratory experiment, a linear regression (PROC REG, SAS Institute 1998) was performed on the total number of ants that died during the 5 d of the experiment, against the log10 of mulch section length. The total number of ant nests and trails found alongside buildings, and the mean number of ants attracted to the baits were analyzed with a mixed model analysis of variance (ANOVA) (PROC MIXED, SAS Institute 1998), with building and building by mulch treated as random effects.

The number of ant nests found around trees was totaled over all dates and compared between mulches using ANOVA (PROC GLM, SAS Institute 1998) after square-root transformation of the data, and subsequent least significant difference (LSD). The presence or absence of nests before the start of the experiment was treated as a factor in the design of the analysis. The number of ants per min trailing on the trees was compared between mulches using ANOVA (PROC GLM, SAS Institute 1998), after square-root transformation of the data.

Results and Discussion

Mortality because of Movement Across Mulch. Ant mortality was positively correlated to mulch section length where sucrose was available only on the end opposite the nest (set B), requiring ants to cross the aromatic cedar mulch to feed (Fig. 1; Table 1). Conversely, mortality and mulch section length were not correlated when ants were able to access the sucrose without having to cross the mulch (set A). At the end of the 5 d experiment, 20–30% of the ants had died in the control (no mulch) treatments of both sets and in all treatments of set A, whereas ≈80% of the ants were dead in the arenas of set B containing 75 cm of mulch (Fig. 1). Low mortality in set A may have occurred because most ants consumed the sucrose near the nest, rather than trailing across the slightly repellent and toxic (Meissner and Silverman 2001) mulch to the distal food source. In set B, greater mortality, together with daily collection of ants at the position opposite the nest (Table 2), indicates that ants were not sufficiently repelled by the mulch to prevent them from walking across it to obtain food (even though mortality was slightly lower at the far end of the longest mulch band compared with the three shorter ones). Similarly, Thorvilson and Rudd (2001) found only a

Table 2. Number (± SEM) of dead ants found on the end opposite the nest in arenas containing aromatic cedar mulch sections of different lengths

	Control	2.5 cm	5 cm	15 cm	30 cm	76 cm
Sucrose on both ends (Set A)	8.9 (2.2)	21.4 (4.5)	16.6 (5.6)	7.1 (3.6)	7.3 (2.3)	5.1 (1.5)
Sucrose on far end only (Set B)	21.8 (4.0)	9.9 (3.0)	32.0 (6.8)	17.4 (4.9)	13.0 (3.3)	7.6 (1.9)

Data averaged over days and replications (n = 16); data include days 1–4 of the experiment.

Table 3. Number of ant nests and trails in aromatic cedar and cypress mulch placed alongside buildings

Mulch	Nests		Trails	
	Mean (± SEM)	N	Mean (± SEM)	N
Aromatic Cedar	0.5 (0.5)a	3	2.6 (0.8)a	3
Cypress	4.3 (1.6)a	3	3.4 (1.0)a	3

Means within a column followed by the same letter are not significantly different (PROC MIXED; $P = 0.05$).

slight repellency of aromatic cedar mulch to the red imported fire ant and hypothesized that the repellency was partially counteracted by the attractiveness of food.

Effect of Mulch Alongside Buildings. Over all sampling dates, only three nests were found in aromatic cedar, compared with 26 nests in cypress mulch. However, the difference in the number of ant nests was not statistically significant (Table 3; df = 1,2; $F = 10.81$; $P = 0.08$). The results can therefore not be considered consistent with the findings of our previous laboratory experiments, where Argentine ants rejected aromatic cedar mulch as a nesting substrate, while they readily accepted cypress mulch (Meissner and Silverman 2001). The first detection of a nest in aromatic cedar mulch occurred 8 wk after the first detection in cypress (Fig. 2). It is not known which of the numerous chemical compounds in aromatic cedar wood are active against the Argentine ant and how stable they are; however, our previous laboratory study demonstrated toxic effects of the mulch on Argentine ants, even after the mulch had been aged outdoors for 140 d (Meissner and Silverman 2001).

Likewise, the number of ant trails leading across the mulch was not significantly different between mulch types (Table 3; df = 1,2; $F = 0.31$; $P = 0.63$). This may have occurred because aromatic cedar mulch did not completely repel ants, as revealed in the above laboratory experiment. Possibly, the magnitude of trailing activity differences may have been obscured by ants trailing below the surface and consequently being less

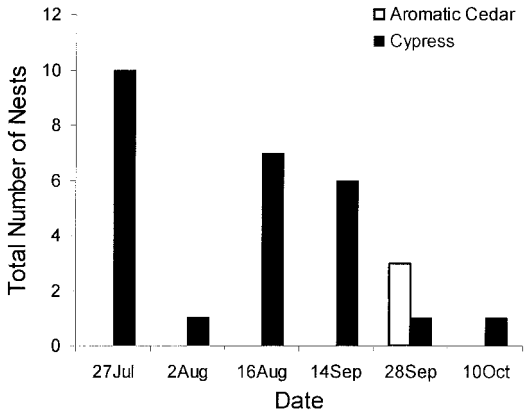

Fig. 2. Total number of Argentine ant nests found on each sampling date in aromatic cedar and cypress mulch alongside buildings.

Table 4. Number of ants on baits placed at two different heights on (or above) aromatic cedar or cypress mulch around buildings

Mulch	Bait Height			
	1 m high		On ground	
	Mean (± SEM)	N	Mean (± SEM)	N
Aromatic cedar	2.8 (0.8)a	39	1.8 (0.5)a	39
Cypress	4.1 (1.1)a	39	1.7 (0.7)a	39

Means within a column followed by the same letter are not significantly different (PROC MIXED; $P = 0.05$).

visible in cypress mulch, while in aromatic cedar mulch ants may minimize exposure by trailing on top of the mulch only. Further research is needed to test this hypothesis.

Regardless of the height of bait placement, the number of ants attracted to the baits was not significantly different between mulch types (Table 4; df = 1,2; $F = 0.51$; $P = 0.55$ for high baits; df = 1,2; $F = 0.03$; $P = 0.88$ for low baits). This again demonstrates that the ants crossed aromatic cedar mulch at least to some degree. However, the ants had to cover only relatively short distances (<0.5m) to gain access to the baits. Because of the generally low number of ants on the baits and minimal trailing activity, we were not able to ascertain whether they traveled longer distances across the mulch or if they had moved predominantly along the building walls and other mulch-free areas.

Effect of Mulch Around Trees. Trees surrounded by aromatic cedar mulch had significantly fewer ant nests than trees surrounded by cypress mulch or the original pine needle mulch (Table 5; ANOVA df = 2,18; $F = 3.75$; $P = 0.02$). Cypress mulch was not significantly different from the pine needle mulch, indicating that replacement of the mulch did not affect ant populations, and that ants recolonized the mulch within a short period of time. Presence or absence of ant nests before the experiment did not influence whether mulch was subsequently colonized (df = 1,18; $F = 0.01$; $P = 0.94$). One week into the experiment, the number of nests in cypress mulch was already greater than before the study, whereas in aromatic cedar mulch the first ant nest was not found until 5 wk into the experiment (Fig. 3).

The number of ants trailing along the tree trunks was significantly different between mulch types (df = 2, 21; $F = 4.93$; $P < 0.02$). Again, cypress mulch did not differ significantly from the original pine straw, whereas aromatic cedar mulch had lower ant numbers

Table 5. Number of nests (totaled over the sampling period) per tree and number of ants trailing on the trunks of trees surrounded by aromatic cedar, cypress, or pine needle mulch

Mulch	Nests		No. ants on trails	
	Mean (± SEM)	N[a]	Mean (± SEM)	N
Aromatic Cedar	0.5 (0.2)	8	37.9 (4.5)	8
Cypress	2.9 (0.61)	8	64.3 (5.7)	8
Pine Straw	2.9 (0.35)	8	56.3 (5.4)	8

[a] Number of trees in each treatment.

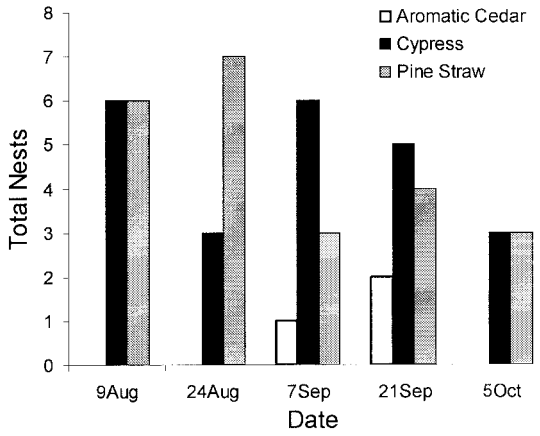

Fig. 3. Total number of Argentine ant nests found on each sampling date in aromatic cedar and cypress mulch, and in undisturbed pine needle mulch (control) around trees.

(Table 5). To a large part, this was probably a result of the absence of nests in the aromatic cedar mulch. However, given that acceptable alternative nesting substrate (e.g., original pine straw) was still surrounding the aromatic cedar mulch, it may also indicate that many workers avoided crossing the mulch.

We suggest that aromatic cedar mulch may reduce the number of Argentine ant nests in certain, well-defined areas, such as in plant beds, alongside buildings, around landscape trees, around picnic tables, and so forth, thereby also reducing ant activity in these areas. Results from our laboratory study suggest that prolonged contact with aromatic cedar mulch is detrimental to Argentine ants (Meissner and Silverman 2001). In situations where availability of nesting sites is limited, aromatic cedar mulch may be used to restrict nesting possibilities even further, thereby decreasing ant population size. Klotz et al. (1997) proposed that the use of repellents, in combination with traditional methods, may lead to more successful ant control in residential areas. We suggest that aromatic cedar mulch may be incorporated into residential ant management programs to complement conventional methods and reduce insecticide input. Additional research is needed to characterize situations where the application of aromatic cedar is most promising, and to test its application in combination with other control methods.

Acknowledgments

We thank Eleanor Spicer for valuable technical assistance, Patricia Alder, Grzegorz Buczkowski, and Gissella Vasquez for reviewing an earlier version of this manuscript, and Cavell Brownie for statistical advice. This study was supported by P&L Bark, Pageland SC and the Blanton J. Whitmire endowment at North Carolina State University.

References Cited

Adams, R. P., C. A. McDaniel, and F. L. Carter. 1988. Termiticidal activities in the heartwood, bark/sapwood and leaves of *Juniperus* species from the United States. Biochem. Syst. Ecol. 16: 453–456.

Appel, A. G., and T. P. Mack. 1989. Repellency of milled aromatic Eastern red cedar to domestic cockroaches (Dictyoptera: Blattellidae and Blattidae). J. Econ. Entomol. 82: 152–155.

Beattie, A. J. 1985. The evolutionary ecology of ant-plant mutualisms. Cambridge University Press, New York.

Bond, W., and P. Slingsby. 1984. Collapse of an ant-plant mutualism: the Argentine ant (*Iridomyrmex humilis*) and myrmecochorous protecaeae. Ecology 65: 1031–1037.

Enomoto, T., S. Ohnishi, Y. Dake, A. Shibano, T. Sakoda, Y. Saitoh, and H. Sogoh. 1999. Environmental control for allergic diseases–avoiding and killing effect on housedustmite by eastern red cedar. [Japanese]. Arerugi 48: 626–631.

Christian, C. E. 2001. Consequences of a biological invasion reveal the importance of mutualism for plant communities. Nature 413: 635–639.

Holway, D. A. 1998. Effect of Argentine ant invasions on ground-dwelling arthropods in northern California riparian woodlands. Oecologia (Berl.) 116: 252–258.

Kennedy, T. A. 1998. Patterns of an invasion of by Argentine ants (*Linepithema humile*) in a riparian corridor and its effects on ant diversity. Am. Midl. Nat. 140: 343–350.

Klotz, J. H., L. Greenberg, H. H. Shorey, and D. F. Williams. 1997. Alternative strategies for ants around homes. J. Agric. Entomol. 14: 249–257.

Laudani, H., and P. H. Clark. 1954. The effects of red, white, and South American cedar chests on the various stages of the webbing clothes moth and the black carpet beetle. J. Econ. Entomol. 47: 1107–1111.

Markin, G. P. 1968. Nest relationship of the Argentine ant, Iridomyrmex humilis, (Hymenoptera: Formicidae). J. Kans. Entomol. Soc. 41: 511–516.

Meissner, H. E., and J. Silverman. 2001. Effects of aromatic cedar mulch on the Argentine ant and the odorous house ant (Hymenoptera: Formicidae). J. Econ. Entomol. 94: 1526–1531.

Messina, F. J. 1981. Plant protection as a consequence of ant-membracid mutualism: interactions on Goldenrod (*Solidago sp.*). Ecology 62: 1433–1440.

Newell, W., and T. C. Barber. 1913. The Argentine ant. Bulletin (United States. Bureau of Entomology); no. 122. Washington, D.C.: U.S. Dep. of Agriculture, Bureau of Entomology, 98 p.

Nonacs, P., and L. M. Dill. 1988. Foraging response of the ant *Lasius pallitarsis* to food sources with associated mortality risk. Insectes soc. 35: 293–303.

Passera, L. 1994. Characteristics of tramp species, pp. 23–43. *In:* D. F. Williams [ed.]. Exotic ants. Westview Press, Boulder, CO.

Rust, M. K., and R. L. Knight. 1990. Controlling Argentine ants in urban situations, pp. 663–670. *In:* R. K. Vander Meer, K. Jaffe, and A. Cedeno [eds.]. Applied Myrmecology—a World Perspective. Westview Press, Boulder, CO.

SAS Institute. 1998. User's manual, version 7.0. SAS Institute, Cary, NC

Scott, E. W., W. S. Abbott, and J. E. Dudley. 1918. Results of experiments with miscellaneous substances against bedbugs, cockroaches, clothes moths and carpet beetles. U.S. Dept. Ag. Bull. 707.

Smith, M. R. 1965. House-infesting ants of the eastern United States: their recognition, biology and importance. U.S. Dept. Agric. Tech. Bull. 1326.

Thorvilson, H., and B. Rudd. 2001. Are landscaping mulches repellent to red imported fire ants? Southwestern Entomologist 26: 195–202.

Vega, S. J., and M. K. Rust. 2000. The Argentine ant—A significant invasive species in agricultural, urban and natural environments. Sociobiology 37: 3–25.

Ward, P. S. 1987. Distribution of the introduced Argentine ant (*Iridomyrmex humilis*) in natural habitats of the lower Sacramento Valley and its effects on the indigenous ant fauna. Hilgardia 55: 1–16.

Received for publication 16 May 2002; accepted 2 December 2002.