

IMPLICATIONS OF RELATIVE ANT ABUNDANCE AND DIVERSITY FOR THE  
MANAGEMENT OF *SOLENOPSIS INVICTA* BUREN USING BROADCAST BAITS

A Dissertation

by

ALEJANDRO ANTONIO CALIXTO SANCHEZ

Submitted to the Office of Graduate Studies of  
Texas A&M University  
in partial fulfillment of the requirements for the degree of

DOCTOR OF PHILOSOPHY

August 2008

Major Subject: Entomology

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## ABSTRACT

Implications of Relative Ant Abundance and Diversity for the Management of  
*Solenopsis invicta* Buren with Broadcast Baits. (August 2008)

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Higher densities of *S. invicta* in the United States relative to South America are explained mainly by the absence of natural enemies and low interspecific competition (IC). Despite advances in *S. invicta* management, broadcast insecticide baits remain as the primary tool for effective control. I studied interspecific interactions of ants and the use of baits on the management of *S. invicta* to test the following hypotheses: 1) relative abundance of native ants increases ~25% for bait treated sites compared to untreated, 2) behavioral dominance by *S. invicta* decreases ~10% in bait treated sites compared to untreated, and, 3) foraging by *S. invicta* on insecticide baits is higher ~10% in low native ant densities areas compared to high densities.

Experiments were conducted on three sites with different densities of native ants (low, medium, high), but with similar densities of *S. invicta*. An enhanced BACI (Before/After-Control/Impact) design was used. Experimental units consisted of 0.4 ha plots. Three treatments were randomly assigned to units and replicated four times; 1) Slow acting bait, 2) Fast acting bait, 3) Untreated Control. Samples and observations were collected for several weeks before/after the treatments to account for temporal

variation and to determine rates of reinvasion. Ants were monitored using pitfalls and food lures. Interspecific competition was determined by applying placebo bait. Results indicate that different management methods did not impact resident ants when they are at low and medium densities and that *S. invicta* is greatly affected; at high native ant density, competition for these baits is observed affecting both natives and *S. invicta*. Reinvasion of *S. invicta* was reduced in areas of low and medium native ant densities previously treated with baits and native ant abundance increased by ~25%. At high native ant densities, reinvasion of *S. invicta* was similar to Control sites. I conclude: 1) properly used, baits can help in recovery of native ant species that then compete with *S. invicta*; however, rote re-treatments may have a negative impact on restored populations. 2) “bait failures” may be due to interspecific competition when initial native ant densities are high. Preliminary management considerations and recommendations are presented.

To Mom (1947-2005).

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## CHAPTER I

### INTRODUCTION

#### *History and Management of Solenopsis invicta in the United States*

*Solenopsis invicta* Buren, native to South America, was accidentally introduced into the United States > 70 years ago through Mobile, Alabama and has since spread throughout the Southern United States infesting more than 130 million ha (Callcott and Collins 1996, Vinson 1997). Two colony forms are found in invaded areas: the monogyne (single queen) and the polygyne (multiple queens) form. Monogyne colonies, characterized by a nest containing a single queen with strongly territorial workers, were predominant for decades following the original invasion in Alabama. In the early 1970's, polygyne colonies were detected and since have become the predominant form (Glancey et al 1973, Porter et al 1991). Each polygyne colony contains many queens and fewer workers than monogyne colonies but polygyne colony densities per ha are double or triple those of monogyne (Porter et al 1997). Foraging territories and workers are shared among related polygyne colonies. *S. invicta* prefers disturbed habitats where they rapidly establish and proliferate. These key factors hinder the management of this invasive species, increasing the cost of management and increasing the risk to humans and animals (Macom and Porter 1996, Wojcik et al 2001)

Management of *S. invicta* traditionally has involved suppression by broad spectrum insecticides, which have caused a dramatic reduction of native ant densities thus accelerating the resurgence and dominance of *S. invicta* (Markin et al 1974, Wojcik et al

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This dissertation follows the style of Ecological Applications.

2001, Carson 2002, Tschinkel 2006). The absence of natural enemies and the lack of strong interspecific competition explain the dominance and higher abundance of *S. invicta* in the United States relative to that in their native South America (Porter et 1988, Porter et al 1991, Porter 1992, Porter et al 1997).

One of the major factors regulating ant densities is interspecific competition (Hölldobler and Wilson 1990). Recent field studies conducted in South America indicate that interspecific competition with other ants appears to be a much stronger factor regulating *S. invicta* densities than mortality ascribed to parasitic flies (Phoridae) (Porter et al 1997, Morrison and Porter 2005, Holway 1999, Feener 2000, LeBrun et al 2007). Relying on these findings and these concepts, improvements in *S. invicta* management practices should include the restoration and preservation of native ants in the United States. This is particularly important for the success of management programs that include the introduction of natural enemies from South America. This interspecific competition appears compatible with biocontrol and is expected to help regulate *S. invicta* densities.

The question that arises from this approach is, are there enough ants (in terms of diversity and abundance) in local assemblages to out-compete *S. invicta*? Several studies have shown that many ant species coexist with *S. invicta* (Helms and Vinson 2001, Morrison 2002). In fact, *S. invicta* affects the densities and distributions of individual species and not species diversity (Morrison 2002, Calixto et al 2007b). Other studies suggest that ant species diversity is greater in similar habitats in South America compared to those in the United States



*S. invicta* research and management programs have included studies of interactions among *S. invicta* and other ant species seeking strategies to limit *S. invicta* impacts on the environment. Research indicates that some ant species are able to eliminate small newly established colonies of *S. invicta* or prevent their establishment by attacking newly mated queens (Nickerson et al 1975, Nichols and Sites 1991, Mottern et al 2004, Rao and Vinson 2004). The majority of these studies involved laboratory-based trials between *S. invicta* and other ant species focusing on one-to-one interactions under controlled conditions rarely if ever observed in the field. These results indicate studies of natural systems on a larger scale, accounting for all the species occurring within the assemblages, through manipulative field experiments are warranted. . This suggests that a slight reduction in *S. invicta* density using other methods may synergize latent competitive capabilities of the resident ant fauna towards the invasive ant (Porter et al 1997) by increasing their niche overlap and increase interspecific competition with *S. invicta*.

#### *Broadcast Insecticide Baits*

Despite advances in management practices and the recent introduction of biological control agents (i.e., phorids, *Thelohania*), pesticides (particularly broadcast baits) remain as the primary tool for *S. invicta* management in the United States (Drees et al 2002, Riggs et al 2002, Barr et al 2005, Aubuchon et al 2006). They are the most cost-effective means of controlling *S. invicta*. Poison baits for *S. invicta* control are typically small granules (~1mm) derived from defatted corn cob and mixed with soybean oil and the active ingredient (an insecticide dissolved in the oil). The most common active

ingredients found in these baits are those carrying metabolic inhibitors, growth regulators and neurotoxins (i.e. hydramethylnon, methoprene, indoxacarb, fipronil). Ant workers are attracted to the bait and carry it back to the colony, where it is fed to the larvae, workers, and queen(s). Although active ingredients have different modes of action, they all serve to break the life cycle of the colony, resulting in its death. Fast-acting short residual baits kill the queen and, to varying degrees, worker ants (Barr et al. 2005). These baits pose very little toxic threat to people and animals as they are broadcast at very low rates (typically 0.6 kg/ha). These baits are not considered selective for *S. invicta* although their effects on non-target ants are relatively unknown. The fast-acting baits (neurotoxins) differ from slower acting insecticide baits (i.e. growth regulators) in their mode of action. Understanding the effects these baits have on the ant community is critical for development of sustainable *S. invicta* management practices in the United States.

#### *Pilot Studies*

A pilot study was conducted in Texas during 2000-2003 to document the impact of broadcast baits on *S. invicta* and non-target species. The manipulative field experiment reduced local densities of *S. invicta* with an Insect Growth Regulator (IGR) bait (Extinguish<sup>TM</sup>, 0.5% s-methoprene, slow active bait) in a managed ecosystem (Harris et al 2003) – a commercial pecan orchard. This study indicated the impact of this bait was greater on *S. invicta*, than on the other ant species. The assemblage was observed to increase in abundance by ~25% in relation to control sites following the reduction of *S. invicta*. The bait treatment benefited numerous native ant species (i.e., *Dorymyrmex*

*flavus*, *Diplorhoptrum molesta*, *Pogonomyrmex barbatus*), resulting in a significant increase in alpha diversity (species turnover). The same study also reported that some of the native ant species still co-existed at high densities with *S. invicta* two years after the last IGR bait treatment (Calixto et al 2007b). One species in particular, *Dorymyrmex flavus* (pyramid ants), showed a significant increase in IGR-treated plots. *D. flavus* was observed strongly interacting with marked *S. invicta* workers and marked *S. invicta* carcasses. Marked remains were later found mixed with much higher densities of unmarked dead *S. invicta* remains on *D. flavus* middens (Calixto et al 2007a). Though direct predation was not observed in the field some laboratory studies suggests *D. flavus* is able to raid small *S. invicta* colonies and to disarticulate workers in the process (Warriner 1998) which explains the larger numbers of remains found on *D. flavus* middens located in areas previously treated with baits.

Density increases of *Pogonomyrmex barbatus* (red harvester ants), an important source of food for the endangered Texas Horned Lizard, were also positively correlated with *D. flavus* increase following reduction in *S. invicta* with IGR bait (Calixto 2007a). Field observations and nest mapping indicate that the two species had similar habitat requirements and they selected similar nesting areas, resulting in a mosaic of *P. barbatus* colonies surrounded by *D. flavus* (Calixto et al 2007a). *S. invicta* have been widely implicated in the disappearance of *P. barbatus* (and indirectly, in the decline of Texas Horned Lizards, *Phrynosoma cornutum*) through direct predation on *P. barbatus* brood and workers (Hook and Porter 1990). These findings suggested that *P. barbatus* may benefit from the close proximity of *D. flavus* nests and their aggressive behavior towards

*S. invicta* in an area treated with IGR bait. Increased diversity was also observed in a community-wide *S. invicta* management project conducted by Texas Cooperative Extension that documented increased species diversity in neighborhoods treated with various bait products (Riggs et al 2002).

Native ants have also been monitored in pastures that have received regular aerial applications of bait blends of Extinguish and Amdro Pro (0.73% hydramethylnon) using different sampling techniques. Native ant abundance was observed to increase, with particular benefits to *D. flavus* and *Diplorhoptrum molesta* (C. L. Barr, A. Calixto and B. Drees, USDA-ARS Area-wide Project internal data).

These studies provide strong evidence suggesting that suppression of *S. invicta* is far more beneficial to native ants than any ill effects to the latter from broadcast baits, contrary to what King and Tschinkel (2006) have suggested in their study in Florida. The use of broadcast baits in particular have been discouraged in areas of relatively low *S. invicta* density and/or high native ant density as a means of conserving native ants (Drees et al 2002, King and Tschinkel 2006, Tschinkel 2006) because such baits were presumed incompatible with native ants.

#### *Biotic and Abiotic Factors Affecting S. invicta Success on Introduced Areas*

Invasive or nonindigenous species are defined as those that have been accidentally or intentionally transported outside their native (historic) range, and spread in their new environment (Levine 2000). Human mediated transport is likely more important to determining ranges, whereas abiotic and abiotic processes (e.g. weather, competitive ability, lack of natural enemies) may be more important for densities of invasive species

(Richardson et al 2000, Colautti and MacIsaac 2004). The general stages of invasion process can be defined as 1) introduction, where the invader is transported from its historic range, 2) establishment, where the invader growth from low densities, 3) spread, where the invasive species exhibit expansion across the landscape, and 4) impact, where the invasive exerts negative, positive effects on resident species (Levine 2000). Two major questions arise regarding this process of invasion: 1) do more diverse assemblages resist invasion? 2) and second, what traits regulate the successful establishment and spread on invaded areas? One possibility that has been suggested is to determine the filters invasive species go through during the invasion process and even look at ecological traits they are introduced into a potential invaded area.

Invasive species, like *S. invicta*, go through many different filters before it is successfully established in their new environments and assembled into an existent community (Hobbs and Norton, Tschinkel 2006). Many ecologists have recognized these filters and the role they may play on developing management strategies tailoring the control of invasives and restoration of species and ecological processes (Hobbs and Norton 2004, Morrison 2008).

Potential invaders begin as propagules residing in a donor region, then pass through a series of filters that may preclude transition to subsequent stages (e.g. establishment, spread). General filters can be defined as 1) propagule pressure, 2) physicochemical requirements of the invader, and 3) community interactions. Determinants may positively (+) or negatively (-) affect the number of propagules that successfully pass through each filter (Colautti and MacIsaac 2004). Hobbs and Norton (2004) recognized

similar filters for restoration programs; Morrison (2008) modified them to apply more directly to animals and plants and appeared to be more concise and applicable to invasive species in contrast to those described by Colauatti and Maclsaac (2004). He defined 1) abiotic and 2) biotic filters. He considered abiotic filters climate, substrate and landscape structure. Biotic filters he considered are competition, predation, propagule availability, mutualism, disturbance, succession and biological legacy. Parasitism and disease are also important filters to consider within the context of *S. invicta* management.

In this study I deal in particular with competition. As with any other invasive species and in the particular case of *S. invicta*, we need to quantify the responses of species to one another and to small changes in the physical environment. Management of *S. invicta* includes designed efforts to manipulate these filters to arrive at a desired species composition and to achieve sustainable management of *S. invicta*. Competition is one of these filters that can be manipulated, possibly using insecticide baits, to achieve sustainable management of *S. invicta* and restore native ant assemblages.

#### *The Role of Competition*

Competition is not the only mechanism that may be invoked to explain patterns of fauna assembly (Kelt and Brown 2004). However the mechanisms involved in how native ants escape insecticidal effects as previously presented are largely undetermined, but appear to be explained by competition. Ant species within an assemblage are often subject to a trade off between exploitative and interference ability that permits species with different foraging strategies to coexist (Holway 1999). There are 3 common

foraging strategies among ants. The first are “opportunists” that arrive first at food sources but are timid and withdraw in the face of interspecific competition. Second are “extirpators”, which often take longer to locate food but recruit in large numbers and aggressively displace other species. Finally, “insinuators” depend on their small size and utilize inconspicuous behavior to collect food while in the presence of other ants (Wilson 1971). *S. invicta* combine fast arrival and massive recruitment becoming, in effect, an opportunistic extirpator and out-compete other predominantly opportunist or extirpator ant species (Porter et al 1988, Camilo and Phillips 1990, Porter and Savignano 1990). Competition in this case is asymmetric, with the advantage going to species able to recruit higher numbers per unit area (Holldobler and Wilson 1990). This scenario leads to linear dominance hierarchies, in which some species that arrive first and take away the limited resources “indirectly” displacing other species not able to compete for the resource, or where dominant species “directly” displace other species at food sources. These two cases are reflective of resource and interference competition (Krebs 2001).

These compensatory mechanisms where resource discovery and dominance are involved appear to define the guidelines for coexistence in local ant communities (Davidson 1998, Holway 1999, Feener 2000) - what Feener (2000) refers to as a “dominance-discovery” trade off. Tilman (1982) emphasized that scientists need to study the mechanisms by which competition operates and the resources that are being utilized. He argued that competition is rare in natural populations and that the few interactions involving resource utilization may not lead to exclusion. Competition appears to be

common through the evolutionary history of communities that has resulted in adaptations (i.e. behavioral shifts) to minimize competitive effects.

In a Tennessee study with the hybrid imported fire ant (*Solenopsis invicta* x *S. richteri*), native ants were able to control food sources better in areas where hybrid fire ants had been suppressed with individual mound treatments (Gibbons 2001). This strongly suggests that *S. invicta*'s rapid and aggressive response to any bait may protect native ants from exposure to the initial treatment independent of toxicity. But, it leaves open the question of whether they would be affected if *S. invicta* were at low densities or absent.

Limited information is available as to how suppression of *S. invicta* benefits native ants and even less is known regarding the role of native ants in resisting *S. invicta* re-invasion after a bait treatment. King and Tschinkel (2006) attempted to address this issue. Through a manipulative field experiment, they explored the effects of fire ant reduction on resident ant abundance and diversity. *S. invicta* was reduced using hot water applied directly to the colony and then observing the response of resident ants to the reduction. Samples were taken during a week in the summer in two years. Results suggest fire ants do not competitively suppress other ant species; however, the study is not well replicated in space and time and appears to be limited to a single week when many ant species may not be active.

Similarly, the toxic effects of insecticides designed for *S. invicta* control cannot necessarily be extrapolated to native ants. Despite this lack of data, the use of *S. invicta* control products has been discouraged in areas with native ant species and an arbitrary



“treatment threshold” of 20 colonies per 0.4 ha (1 acre) is often used to trigger treatments (Drees et al 2002). Research is needed to define this threshold with biological, economic, and/or human nuisance parameters.

*S. invicta* management strategies must be tailored to human expectations for use of the infested area. Broadcast baits are the most effective, least expensive and least toxic means of *S. invicta* control in most situations (Barr et al 2005). Broadcast baits offer great flexibility in developing tailored strategies because they vary in type of toxin, speed of action, residual activity, effects on non-target species, and mode of action. Regardless of overall *S. invicta* density, a single colony in the “wrong” place (such as in the breeding site of an endangered species) can cause substantial damage, thus necessitating the need for preventive treatments, most typically applied as broadcast baits. Yet, baits are not recommended for use in areas of low *S. invicta* density or native ant presence, despite there being a dearth of data from the field to support these recommendations. Careful examination of commercially-available *S. invicta* control products in the field is needed to document their actual effects, and then to explore development of strategies tailored to human and wildlife needs.

The study sites in this project are represented by unimproved grasslands with scattered woods under low intensity grazing. The intensive sampling proposed will provide a database for *S. invicta*, native ants, and other macroscopic arthropods’ responses and interactions to a variety of different baits. I expect knowledge gained in this study to allow tailoring a range of management strategies for *S. invicta* and for the restoration/conservation of native ants in Texas.

This study investigates, through field manipulative experimentation, the effects on the relative abundance and diversity of native ants to the application of prescribed baits (fast and slow active ingredients) and reduction of *S. invicta*. The rate of competition (dominance and discovery) of broadcast baits during the application, and the impact of reinvasion of *S. invicta* in areas of high, medium and low density of native ants treated with baits will be evaluated in typical Post Oak Savannahs in Central Texas. I attempt to use the Adaptive Management approach. This uses management practices as experiments which must be monitored carefully to ascertain if goals were met and to identify errors in understanding the dynamics of the systems managed. In this approach actual mechanisms and effect to management actions are compared to our preexisting information of the system, then corrected and implemented (Walters 1986). With this approach new *S. invicta* management strategies will be developed tailoring human needs by determining attributes of particular poison baits and judiciously combining the best management practices with interspecific interactions from native ants. Ideally, *S. invicta* can be sustained at low densities for extended periods reducing the costs of management and frequency of re-treatments for the control of this species in vulnerable areas in Texas.

The objectives of this investigation are 1) determine the effect of different broadcast baits on native ant diversity and relative abundance in Post Oak Savannahs in Central Texas; 2) determine if different relative abundances of native ants slows and/or prevent re-invasion of *S. invicta* into treated areas with broadcast baits; 3) determine the mechanisms for access and removal of broadcast baits by *S. invicta* and native ants to

develop treatment recommendations to improve the use of these products against *S. invicta* and/or minimize effects on native ants; and 4) gather baseline data on other arthropods to determine whether secondary effects occur due to the alteration of the ant assemblage in the rangeland ecosystem.

## CHAPTER II

### MATERIALS AND METHODS

#### *Study Sites*

This study was conducted at three sites located on private lands in Central Texas (Fig. 1). Two of these sites are located in (Coryell Co.) 193 kilometers NW of College Station (“Pruitt”: 31°30'19.33"N, 97°43'32.32"W and; “C3”: 31°25'36.87"N, 97°36'44.25"W). These sites are part of the Leon River Restoration Program (Institute of Renewable Natural Resources, Texas A&M University). The third site is located in (Burlinson Co.), 20 kilometers SW of College Station (“Barr”: 30°26'2.22"N; 96°30'41.12"W). The habitats consist primarily of grazed post-oak savannah with areas of dense brush near seasonal creeks. The sites were screened to detect absence or presence of phorid flies (*Pseudacteon spp.*) and *Thelohania solenopsae* (a natural disease occurring statewide that infects *S. invicta* colonies). Phorid flies have been released on several sites in Texas and are spreading off the original release sites (Gilbert et al 2008). Since the impact of these introduced organisms remains unclear, sites were located out of the expansion range of phorid flies to avoid introducing potential variation. *T. solenopsae* has been found across most of Texas and the persistence of the infection appears constant through the state (Mitchell et al 2006). Therefore, a homogeneous effect is expected across sites. All sites in this study were positive for *T. solenopsae* but free of *Pseudacteon* flies.

### *Research Hypotheses*

These hypotheses were established based on findings by Calixto et al (2007b). Effect size represents the change in density associated with the bait treatment and the reduction of *S. invicta*.

*Hypothesis 1.* Abundance of native ants increases by at least 25% (Effect Size  $\geq 0.6$ ) for bait treated sites compared to untreated.

*Hypothesis 2.* Behavioral dominance by *S. invicta* decreases by at least 10% (Effect Size  $\leq -0.2$ ) in bait treated sites compared to untreated.

*Hypothesis 3.* Foraging by *S. invicta* on insecticide baits is higher by at least 10% (Effect Size  $\geq 0.2$ ) in low native ant densities areas compared to high densities.

### *Experimental Design*

Experiments were conducted using an enhanced BACI-P (Before/After-Control/Impact-Paired) design, which had two sets of sampling replicates in time (1 before and 1 after the impact) with measurements of biological and environmental data in area by time factorial design. A pulse experiment was used to measure the response to a single treatment application (resilience). Treatments were applied on 15 May 2006 and 17 June 2007 respectively. All three sites were sampled from March through October during the 2006 and 2007 seasons, with March-May sampling being the set of replicates before the impact and June-October the set of replicates after the impact.

*Initial densities of S. invicta and native ants.* The three sites were rigorously sampled by using pitfall traps and food lures to determine the relative abundance and behavioral dominance of both *S. invicta* and native ant species. Sixty pitfall traps and 96 food lures

(beef/chicken wieners) were deployed on each one of the sites on areas where eventually the plots were going to be located. Sites were sampled for up to seven weeks during 2006. Data indicated the three sites had similar relative abundance and behavioral dominance of *S. invicta*, but differed in the abundance of native ants ( $P < 0.05$ , Linear Mixed Model, Tukey Post hoc analysis). Sites were then blocked by native ant abundance as low (“Pruitt”), medium (“Barr”) and high (“C3”) (see Fig. 2).

*Experimental plots.* Field experiments used 0.5 ha plots. Plots were created with the software ArcGIS (ESRI) and using the Xtools extension. This extension was used to create a grid on the sites map; cells with similar characteristics (i.e. soil and vegetation characteristics) were grouped to reduce the source of variation. Each cell represented a 64x64m square plot. Final plots were then selected at random from the grids, and established in the field using a Trimble GPS (sub meter accuracy), and marked. A buffer of at least ~35m was left between plots to lessen the possibility of treatment effects between plots (following Drees and Barr 1992, Martin et al 1998).

*Treatments.* A completely randomized design was used to assign the treatments to the plots (experimental units). The treatments consisted of; 1) a slow acting bait, (IGR - Extinguish, 0.5% s-methoprene, Wellmark International, Dallas, TX 75234), 2) a fast acting bait (Advion, 0.045% indoxacarb, DuPont Professional Products, Newark, DE 19714), and 3) untreated controls. Treatments were applied at a rate of 0.68kg/ha using a Herd seeder (Model GT-77 ATV - Herd Seeder Co. Inc.) mounted on a utility vehicle (John Deere Gator TH 4x4, Deere & Company). Treatments were applied on May 16, 2006 and June 7, 2007 on “Barr” and May 17, 2006 and June 6, 2007 on “C3” and

“Pruitt”. All treatments were applied during the morning (between 8-11 am) and with temperatures lower than 28°C. Treatments were replicated four times. The minimum number of replications needed to detect differences was determined using power analysis based on data collected in previous studies (Calixto 2004) (Power= 0.80;  $\alpha$ = 0.05; Effect Size = ~25% increase). This analysis suggested the use of six or more replicates per treatment. However, due to constraints in space and the limited availability of personnel, four replications were used. Observations in time were used to increase the scale and improve the inferences that could be made from the results.

### *Sampling Methods*

*Relative abundance and behavioral dominance.* Two complementary sampling techniques were used in each experimental plot at each study site. The first sampling technique consisted of pitfall traps to estimate the relative abundance and species composition of ground active ants (Bestelmeyer et al 2000). Traps consisted of a 120 ml plastic cup filled with propylene glycol (commercial antifreeze). Traps with lids were set in the field and opened 48 hours later to minimize “digging in” effects (Greenslade 1973). Five traps were used per plot. Traps were distributed uniformly in the experimental unit (Systematic sampling).

The second sampling technique consisted of food lures (“hot dogs”) (Bestelmeyer et al 2000) used to determine relative abundance and behavioral dominance of ant species. Eight lures were distributed uniformly through the experimental unit (Systematic sampling) in each plot. Lures were inspected after one hour and the number of ants

observed on each slice (following Pereira and Porter 2005). Sampling dates for both pitfalls and food lures are presented in Table 1.

*Competition for insecticide baits.* In each plot, treated and untreated during and after treatment in all field trials, placebo bait (containing soybean oil but lacking active ingredient) was broadcast at 0.68kg/ha using a hand-held rotary spreader. The number of ants foraging on bait particles on the ground inside quadrats were then recorded (Bestelmeyer et al 2000). These quadrats consisted of ½-inch diameter PVC pipe frames, 25x25 cm<sup>2</sup> square. Ten of these units were deployed and distributed uniformly through the experimental unit (systematic sampling). Ants were recorded every 15 minutes for 90 minutes or until the last granule was observed removed. These observations examined how non-target species access baits under a range of conditions vis a vis *S. invicta*.

*Environmental data.* Data loggers (HOBO, Onset Computer Corporation) were placed at each of the study sites. Data on daily rainfall, temperature and humidity were recorded for the entire duration of the study.

#### *Data Analysis*

*Impacts of treatments.* Data were analyzed separately in three different ways, each addressing the hypotheses presented above. I estimated Effect Sizes (ES) (Cohen's *d*) and 95% confidence intervals for the data collected by pitfall traps and food lures. These effect sizes are standardized measures that express changes in density associated with the treatments and, therefore, the magnitude of a treatment effect (Cohen 1988, Scheiner and Gurevitch 2001).

$$d = M_1 - M_2 / \sigma_{\text{pooled}}$$



$$\sigma_{\text{pooled}} = \sqrt{[(\sigma_1^2 + \sigma_2^2) / 2]}$$

Cohen (1988) defined  $d$  as the difference between the means,  $M_1 - M_2$ , divided by standard deviation,  $\sigma$ , of either group. Cohen argued that the standard deviation of either group could be used when the variances of the two groups are homogeneous. In meta-analysis the two groups are considered to be the experimental and control groups. By convention the subtraction,  $M_1 - M_2$ , is done so that the difference is positive if it is in the direction of *improvement* or in the predicted direction and negative if in the direction of *deterioration* or opposite to the predicted direction.

The resulting ES and 95% CI were then plotted to illustrate the biological effects (positive, neutral or negative) and statistical significance observed for each experiment (Scheiner and Gurevitch 2001). The software Metawin 2.0 was used to perform these analyses.

I analyzed the data obtained from the different sampling methods using the Linear Mixed Model (LMM) (Repeated Measures - Type III sum of squares and diagonal repeated covariance) procedure. In this analysis, “plot” was the random factor and “treatment” and “week” were the fixed factors in the model. This analysis allows for unsystematic variability of the data and provides greater power to detect effects. In this method, the covariance of errors is estimated and then used to constrain the error covariance matrix to derive the mean squares estimates of the effects. This method provides a way for modeling error structures among the repeated dependent variables and avoids Type I errors, it is also used for unbalanced sets of data. The statistical

package SPSS 15.0 (SPSS Inc. 2007) was used to perform these analyses (values significantly different when  $P < 0.05$ ).

*Impact of treatments on diversity.* I estimated raw species richness for data collected from pitfall traps and compared them among treatments using LMM. In addition I used a simple estimator of the absolute number of species in the assemblage based on the number of rare species in a sample; this estimator is known as Chao 2 (Colwell and Coddington 1994), and is expressed by the formula:

$$S_{\text{Chao 2}} = S_{\text{obs}} + \frac{Q_1^2}{2Q_2}$$

Where  $Q_1$  = the number of species that occur in one sample only (singletons) and  $Q_2$  = the number of species that occur in two samples (doubletons). Species richness was compared among treatments to detect impacts of treatments on diversity. Rarefaction curves were generated by random re-orderings using the program EstimateS (Colwell 2006). Numbers of species projected by the analysis are reported accompanied by 95% CI and an estimation of sampling efficiency.

*Species co-occurrence.* I used a null model to analyze the co-occurrence structure of ants in treated vs untreated areas. Null models are statistical methods and computer simulations used to analyze patterns in nature (Gotelli 2000). In this case, I examined species co-occurrence patterns to test whether ant communities among the treatments are non-random assemblages (against a null hypothesis that they are randomly assembled) and whether they are predicted by competition with the invasive species. I used pitfall trap data, because they reflected community structure. I tested the data collected against

a null model asking whether there were significant differences in the co-occurrence patterns observed for native ants in treated vs untreated sites. *S. invicta* was not included because the presence of *S. invicta* was used to designate treatments.

Null models tests whether the mean co-occurrence index among treatments is larger or smaller than expected by chance. It also tests the variance of the co-occurrence index among treatments. An unusually large variance would mean that the treatments differ significantly from one another in their levels of co-occurrence: some treatments have species with high levels of co-occurrence and the other has species with low levels of co-occurrence. An unusually small variance would mean that treatments are similar to one another in the level of co-occurrence observed. A random result for the variance means that the level of co-occurrence among treatments is about what would be expected if the species were assigned randomly to different treatments (Gotelli 2000).

## CHAPTER III

### RESULTS

#### *Species Richness*

A total of 675,422 ants and 34 species were collected for the entire study using pitfall traps. During the sampling period of 2006, 121,679; 41,256 and, 268,605 ants were collected at “Pruitt”, “Barr” and “C3” respectively. For the sampling period of 2007, 98,210; 43,990 and, 101,682 ants were collected at “Pruitt”, “Barr” and “C3” respectively. By using the two sampling methods (pitfall traps and food lures), 17 species were found for “Pruitt” (Tables 2 and 3), 22 species for “Barr” (Tables 4 and 5) and 27 species for “C3” (Tables 6 and 7). The most abundant species sampled for all sites were *Solenopsis invicta* Buren, *Forelius pruinosus* (Roger), *Paratrechina terricola* (Buckley), *Monomorium minimum* (Buckley) and *Dorymyrmex flavus* McCook.

Results from pitfall traps for 2006 and 2007 showed no significant differences among treatments before and after the bait application ( $P > 0.05$ , LMM) (Figs. 3 and 4). Results of these analyses for all sites and sampling techniques are presented in Tables 8 and 9. Sample-based rarefaction curves for pooled treatment data for “Pruitt” projected 27 species (95% CI: 16.67-108.94) and 17 species (95% CI: 13.56-41.72) for Methoprene treatments for years 2006 and 2007 respectively, and 18 (95% CI: 16.35-41.46) and 17 (95% CI: 16.13-26.02) for Indoxacarb treatments for years 2006 and 2007 respectively, and, 16 (95% CI: 15.07-29.17) and 15 (95% CI: 13.18-35.13) for untreated Control treatments for years 2006 and 2007 respectively.

For “Barr,” sample-based rarefaction curves projected 31 species (95% CI: 20.26-85.59) and 23 species (95% CI: 15.99-79.82) for Methoprene treatments for years 2006 and 2007 respectively, 16 (95% CI: 15.07-29.16) and 26 (95% CI: 18.99-82.82) for Indoxacarb treatments for years 2006 and 2007 respectively, and, 17 (95% CI: 17.56-24.8) and 24 (95% CI: 18.57-58.53) for untreated Control treatments for years 2006 and 2007 respectively.

For “C3”, sample-based rarefaction curves projected 18 species (95% CI: 18.06-25.8) and 21 species (95% CI: 21.26-86.63) for Methoprene treatments for years 2006 and 2007 respectively, 17 (95% CI: 17.01-20.53) and 32.96 (95% CI: 21.26-86.63) for Indoxacarb treatments for years 2006 and 2007 respectively, and, 20 (95% CI: 19.18-31.44) and 22 (95% CI: 21.26-86.63) for untreated Control treatments for years 2006 and 2007 respectively.

The shape of the curves during 2006 showed that sampling in “Pruitt” and “Barr” accumulated species much more quickly in Indoxacarb and Control treatments compared to Methoprene. At “C3” species appear to accumulate similarly among the three treatments. Overlap of 95% CI indicates treatments among the three sites project similar numbers of species and similar shaped rarefaction curves (Fig. 5).

For 2007, the shape of the curves revealed in “Pruitt” showed similar accumulation of species as in 2006. “Barr” accumulated species less quickly, as it appeared not to reach a plateau in the accumulation of species. In “C3,” Methoprene and untreated Control produced similar accumulation curves as they reached a plateau more quickly than the Indoxacarb treatment. As in 2006 and based on overlap of 95% CI, treatments among the

three sites projected similar numbers of species and similar shaped rarefaction curves (Fig. 6).

#### *Species Co-occurrence*

Analysis of co-occurrence among treatments in the absence of competition by *S. invicta* indicated that the community structure is very similar. Native ant species do not appear to co-occur significantly more or less (small C-score) among untreated areas than among sites that have been treated for *S. invicta* (Table 10). These results suggest community structure is predicted by chance and not by any biological interaction, in this case, competition.

#### *Impact of Bait Treatments on Native Ant's Relative Abundance and S. invicta*

##### *Dominance*

*Relative abundance.* Data from pitfall traps for “Pruitt” during 2006 showed a reduction in abundance of *S. invicta* in both bait treatments compared to untreated Controls following the bait application (Fig. 7). However the LMM analysis showed no significance differences among treatments (Table 11) but Effect Sizes (ES) were closed to the 10% reduction proposed on the original hypotheses (Fig. 8). Native ants at this site exhibit at slight increase in abundance (near 25%, close to the ES proposed on the hypotheses) however the LMM analysis did not show statistical differences. Numbers in some instances were too low to perform the LLM analyses.

Data produced for “Barr” for the same year showed a significant reduction of *S. invicta* due to the bait treatments (Table 12). Relative abundance remained similar after the bait treatment (Fig. 9). ES also showed the magnitude of the bait impact on *S.*

*invicta*, especially by Indoxacarb, which exhibited a fast action. A slight reduction was observed in native ants for this particular treatment after the application (Fig. 10).

For “C3”, the bait treatment had a significant impact on *S. invicta* abundance (Table 13). Native ants were also observed increasing in abundance especially in those areas treated with Methoprene (Fig. 11). However, LMM showed no significance differences in the same year. ES indicated the magnitude of the impact of the bait on *S. invicta* whereas native ants appeared unaffected (Fig. 12).

For year 2007, “Pruitt” showed significant differences among *S. invicta* abundance due to the bait treatments (Table 14) while native ants exhibited a significant increase compared to Control (Fig.13). ES indicated the Methoprene exhibited better control of *S. invicta* compared and a positive response by the native ants (Fig. 14).

For the same year, “Barr” presented a reduction in the abundance of *S. invicta*, however the LMM indicated otherwise (Table 15). ES however, showed that both Methoprene and Indoxacarb appeared to have contributed to the reduction in *S. invicta* unlike the interpretation provided by the LMM (Fig. 15). Based on the LMM and also supported by the ES (Fig. 16), native ants do not appear to be adversely affected by the treatments.

“C3” for 2007 showed significant reduction in *S. invicta* relative abundance due to both bait treatments (Table 16). Native ants exhibited significant increases in bait treated areas (Fig. 17). ES consistently showed the negative impact of bait treatment on *S. invicta* and showed no apparent changes in native ant abundance (Fig. 18).

Figures 19 and 20 present a summary of the estimated ES for both years and all the sites generated using the pitfall trap data. In summary, pitfall trap data for 2006 showed no change due to bait treatments in *S. invicta* abundance nor were native ants negatively impacted. In fact, a slight increase was observed at “Pruitt” near the ES proposed in the original hypotheses. For “Barr” and “C3,” *S. invicta* abundance of workers appeared to be slightly impacted, and a slight reduction in natives was observed, though not enough to be significant. For 2007, the situation was similar, however native ants were slightly reduced (but not significantly).

*Relative behavioral dominance.* Impact of bait treatment and native ant presence on discovery and dominance of resources by *S. invicta* was measured using food lures. For the year 2006 at “Pruitt,” the bait treatment significantly reduced foraging and dominance of *S. invicta* (Table 11). Native ants were observed more at food lures (Fig. 21), but numbers were not significantly higher compared to Control based on the LMM analysis. ES consistently shows an impact of bait treatments on *S. invicta* and, contrary to those results yielded by the LMM, native ants appeared to increase and dominate food lures in areas where bait treatments were applied (Fig. 22).

At “Barr,” dominance of *S. invicta* was significantly reduced (Table 12). Native ants in this site appear to also increase and dominate the food lures in bait treated areas (Fig. 23). As in “Pruitt,” the LMM did not show significant differences, however careful analysis using ES shows a dramatic and significant dominance by native ants (Fig. 24).

“C3” did not show significant reduction of dominance of resources by *S. invicta* (Table 13). Native ants also appeared unaffected by the treatments (Fig. 25). ES were



consistent with these results. The impact of the treatment did not achieve good control of the invasive however native ants appeared slightly affected (Fig. 26).

For the year 2007, “Pruitt” showed significant reduction of *S. invicta* dominance on both bait-treated plots (Table 14). Native ants also significantly increased in dominance in bait treated plots (Fig. 27). ES were also consistent with the impact on *S. invicta* dominance and showed a slight increase of native ants at food lures (Fig. 28).

“Barr” also exhibited significant reduction of *S. invicta* at food lures (Table 15), with native ants also more common at food lures in plots that were treated with baits (Fig. 29). ES estimations were also consistent with the reduction of *S. invicta* and the increase of dominance at lures by native ants (Fig. 30) in 2007.

The bait treatments did not significantly reduce the dominance of *S. invicta* at “C3” (Table 16) in 2007. To the contrary, foragers of the invasive appear more active in bait treated plots. Native ants were significantly more active in Control areas compared to bait treated plots where they tended to dominate food lures – an indication the bait treatment had the greatest impact on the native assemblage (Fig. 31). ES reflected that, in fact, *S. invicta* was not altered by the bait treatment and remained active in those plots, whereas natives appeared to be affected by the bait treatments (Fig. 32)

Figures 33 and 34 present a summary of the estimated ES for all years and sites generated using the food lure data. In summary, food lure data for 2006 showed significant impacts by baits on dominance and relative abundance of *S. invicta* at low and medium native ant densities (“Pruitt” and “Barr” respectively). At “C3”, the site with higher native ant abundances, *S. invicta* appears unaffected by the Indoxacarb

treatment but shows an impact from the Methoprene. Native ants also showed decreases in dominance at food lures especially in those areas treated with Methoprene. For 2007, the bait treatments exhibited a similar efficacy to those observed during 2006 for the sites with low and medium native ant densities (“Pruitt” and “Barr”). On the same sites, native ants exhibit increases in dominance near the ES proposed in the original hypothesis. For “C3,” the bait treatment definitively did not reduce dominance of *S. invicta*, but it did affect foraging and abundance of native ants.

#### *Competition for Bait Insecticides*

During the spring of 2006, the placebo baits in all three sites and in all experimental plots were rapidly discovered and removed by *S. invicta* (Fig. 35), most bait granules being gone in less than two hours. Figures 36-38 shows how *S. invicta* rapidly removed those bait particles. No significant differences were found at this point in the number of *S. invicta* foragers observed at quadrats among the experimental plots (Tables 11-13). Individuals of native ant species were not frequently observed at quadrants and only three species (*S. invicta*, *F. pruinosus* and *M. minimum*) were observed attracted to the bait particles (Tables 17-19).

After the fall insecticide application, frame evaluations indicated more native ant foragers collecting the placebo particles than *S. invicta* (Fig. 35). A shift in discovery and removal was observed in areas where *S. invicta* has been reduced with baits. Nevertheless, *S. invicta* remained, in some instances, dominant at, while other ant species appeared foraging and collecting these baits (Figs. 36-38).

For 2007, a similar situation was found. *S. invicta* quickly discovered and removed the grits before the insecticide bait application (Fig. 39) and, as in 2006, *S. invicta* appeared more actively discovering the particles in areas of low native ant density. At high native ant densities, particle discovery was distributed among *S. invicta* and some native ant species (Figs. 40-42). Following the bait application and the reduction of *S. invicta*, native ants appeared to discover and collect the particles more often. For 2007, *S. invicta* appeared to collect particles less frequently in previously Methoprene treated areas, but the direct effect remains unclear.

*Impact of Baits and S. invicta on Relative Insect Abundance and Taxa Diversity*

Preliminary data obtained using pitfall traps during 2006 indicated that neither bait insecticides had a significant effect on relative insect abundance for any of the sites and proportions remained relatively similar before and after the bait application (Fig. 43). The average number of taxa found in samples appeared to remain constant and was similar before and after the bait application. No significant statistical differences were found both on relative abundance and taxa diversity among the three treatments. Linear Mixed Models results for effects of treatments on the relative abundance and taxa diversity of insects during 2006 are presented in Table 20.

## CHAPTER IV

### DISCUSSION AND CONCLUSIONS

#### *Ant Species Diversity and Co-occurrence*

Ant species found in this study are fairly consistent with those reported coexisting with *S. invicta* in other studies (Helms and Vinson 2001, King and Tschinkel 2006, Calixto et al 2007b). More importantly, this study shows that management of *S. invicta* using insecticide baits does not have a significant impact on species richness and abundance. No species suffered local extinction among the treatments and the number of species projected for each method was relatively close to those observed through the sampling periods. The ants of Texas have been surveyed thoroughly (O’Keefe et al 2002), so species patterns can be evaluated more accurately in the context of species co-occurrence in relation to the invasive *S. invicta*. Intensive sampling at all three sites captured 34 species of ants (almost 12% of all ants reported for Texas (O’Keefe et al 2000)).

Species richness (Chao2) for all sites and treatments did not exhibit a “hump-shaped” pattern observed for disturbed habitats (Rosenzweig and Abramsky 1993). This is particularly unusual considering these Post Oak Savannahs are used for grazing and are subject to disturbance. The pattern that was observed among the three treatments for the two years possibly indicates that the ant assemblage was symmetrically disturbed and responded similarly. Another type of disturbance that was not reflected in the rarefaction curves was the bait application and the reduction of *S. invicta*. These findings are consistent with those reported by King and Tschinkel (2006) and Tschinkel (2006)

where they suggested a reduction of *S. invicta* does not affect ant diversity. In fact, among all treatments, before and after the bait “impact” raw species richness remained similar. *S. invicta* has been considered as the cause of a loss in insect diversity, especially in the ant mosaic (Porter and Savignano 1990). More recent studies showed otherwise (Morrison and Porter 2003). *S. invicta* does not appear to dramatically impact other ants and insects and some are benefitted by the presence of *S. invicta*.

This study shows that “community stability” between *S. invicta* and native ants appears to occur. Results from the null model did show that community patterns are equally predicted in the absence or presence of *S. invicta*. This stability may be mediated by the allocation of resources and how different species access those resources. Some species may have evolved mechanisms to avoid competition, escaping in space and time or developing behavioral strategies that secure their survival (fitness) (Krebs 2001).

Species richness should be highest in favorable localities where competitive exclusion (if it occurs) is reduced by infrequent disturbance events or mildly stressful conditions (Rosenzweig and Abramsky 1993). Whether this applies to this particular study remains unclear.

Finally, the small variances observed among the C-score (Table 10) means that treatments are similar to one another in the level of co-occurrence observed. A random result for the variance indicated that the level of co-occurrence among treatments is about what would be expected if the species were assigned randomly to different treatments, these findings contradict those studies that suggested *S. invicta* and other invasive ant species causes community disassembly (Gotelli and Arnett 2000, Sanders et

al 2003, Calixto et al 2007b). In these studies, sites without the invasive species ant assemblages exhibit significant species segregation, consistent with competitive dynamics. In sites with the invasive ant, ant assemblages appear random or weakly aggregated in species co-occurrence. These results indicate that invasive species reduce biodiversity but rapidly disassemble communities. The findings from my study show strong species co-occurrence in both reduced and untreated areas, indicating *S. invicta* does not disassemble the ant assemblage nor reduced the number of species in this particular ecosystem. Several hypotheses can be drawn from these results, 1) coexistence with the invasive ant has been common but the limited scale of natural and manipulative experiments have led to the conclusion that *S. invicta* excludes resident species, 2) resident ant assemblages have evolved physiological and behavioral mechanisms to avoid competition and predation from *S. invicta* considering this species has been in contact with indigenous species for more than 70 years since its original introduction, evolutionary processes at local levels may be undergoing, 3) the different filters present at introduced locations have level down the net effects of this invasive on population levels of other ant species, and 4) a combinations of all these factor its occurring.

#### *Impact of Bait Treatments on S. invicta Dominance and Native Ants*

At the beginning, I hypothesized that abundances of native ants were expected to increase near 25% in bait treated areas compared to untreated. These differences were obtained from studies conducted by Calixto et al (2007b). Pitfall trap data and especially food lure data (behavioral dominance) demonstrated that, in some instances, the experiments achieved the effect proposed in the original hypothesis, indicating that the

bait application does not exert significant impact on native ant abundance. Pitfall trap data for the two years was consistent in showing no impact of bait treatments on natives. But, it did not provide conclusive evidence that an increased abundance occurs, as was observed in the study conducted by Calixto et al (2007b). However, in that study, the number of traps used for sampling ants was significantly lower compared to this study. The data however appears to have some characteristic “noise” and these traps are better at describing community structure and diversity than abundance (Bestelmeyer et al 2000). Food lures, though, showed marked dominance by *S. invicta* which consistently discovered and dominated food lures, as is observed at natural resources (Tschinkel 2006, LeBrun et al 2007, Calcaterra et al 2008). *S. invicta*, before bait treatment at low and medium native ant densities, appeared to rapidly recruit to food lures and dominate the resource, thus displacing other ant species. Following the bait treatments, *S. invicta* does not appear to be the dominant species and a shift in hierarchy is observed in ants of similar size and with the ability to produce defense mechanisms (Holldobler and Wilson 1990, Tschinkel 2006). Several ant species, especially *Forelius pruinosus*, tended to dominate the resources when *S. invicta* was reduced with baits. These results are consistent with those obtained by Calixto et al (2007b) where behavioral dominance by other ant species occurs when *S. invicta* is reduced. This is also in agreement with the theory of “discovery-dominance trade off” proposed by Feener (2000) and covered by Holldobler and Wilson (1990). In this case, *S. invicta* appeared to be reduced to levels where other ant species were able to compete with them and exhibit similar community structure found in their original territories in South America (Macom and Porter 1996).

This presents an interesting scenario for the management of *S. invicta* - inducing interspecific competition by selectively reducing *S. invicta* and allowing increase in fitness of native species. More studies are needed to investigate further the impact on fitness of *S. invicta* under this management scenario.

#### *Competition for Bait Insecticides*

The third and last hypothesis presented assumed that if *S. invicta* is the dominant species, with more colonies per unit area and with larger colonies than resident ant fauna (Tschinkel et al 1995), that the chances for them to encounter insecticide baits is greater than those for native ants. In this study I found that *S. invicta* discovers and removes the particles more quickly than resident ant species and that, in most of the cases, no more than four species access these particles. Based on these results, I argue that, given the ability of *S. invicta* to quickly discover and dominate resources, management of this species with insecticide baits under several environmental conditions is the preferred means of control. The trade off is that at relative high native ant abundance, interspecific competition may occur. Therefore, the “resources” in this case, the bait particles, are allocated to both the invasive and resident ants, thus causing some degree of mortality to a few native ant species.

Interspecific competition among ants occurs most frequently among similar (size, morphology, life history) species (Holldobler and Wilson 1990). Given these circumstances, it is expected that out of the ant mosaic found in Post Oak Savannahs in Texas that just a few species may access these baits and be adversely affected. In this study, the most common ant species attending the particles (exclusive of *S. invicta*) were



*Forelius pruinosus* and *Monomorium minimum*, two species known to engage in strong interspecific competition with *S. invicta* and also known to attack and raid small *S. invicta* colonies and kill newly mated queens (Nickerson et al 1975, Nichols and Sites 1991, Mottern et al 2004, Rao and Vinson 2004). Other ant species within the assemblage are not expected to be affected by the bait treatment. These mechanisms of interspecific competition (interference and resource) at play in this ecological scenario support the well know theory of niche specialization, which results in differences in relative survival across niches by different ant species and in the rates of production of different niches (Krebs 2001).

The use of insecticide baits has created a large controversy in the US. Many believe the use of these products harm resident ant fauna and that the active ingredients enter the food chain affecting other non-ant species (King and Tschinkel 2006, Tschinkel 2006). The information in this regard is very limited as few sound, intensive manipulative experiments have been conducted. This study shows bait treatments (applied at the rate indicated by the product label and authorized by the Environmental Protection Agency, EPA) have little effect on native ants when native ants are at low and medium densities and *S. invicta* is present in significantly higher numbers.

#### *Impact of Baits and S. invicta on Relative Insect Abundance and Taxa Diversity*

*S. invicta* is an omnivorous predator and scavenger, with insects comprising most of their diet (Killion et al 1995). The extent of the impacts of *S. invicta* on native faunal communities is not yet known. *S. invicta* have been shown to reduce the diversity of arthropod communities and to have negatively affected other invertebrate fauna (Lofgren

1986, Wojcik et al 2001). It has long has been debated whether insecticide baits harm non-ant species and whether *S. invicta* exerts an impact on non-ant communities. Invasive species are known not only to displace closely related species but also to outcompete other species and prey upon them. The impact of these two factors is relatively unknown (Tschinkel 2006). I attempted to provide baseline data to document both the impact of bait insecticides and *S. invicta* on insect abundance and diversity of ant taxa.

Results reported in this study indicate that bait insecticides do not appear to negatively affect insect abundance and insect taxa. This was consistently observed for all the sites of this study. No evidence was found to indicate that the reduction of *S. invicta* using these products significantly increased the abundance of insects. The bait application, given that the active ingredients s-methoprene and indoxacarb are very volatile and longer exposure to warm temperature and moisture appears to dissipate the effect (Moncada 2003, Csondes 2004), and the fact that most of the grits are probably collected by ants, does not appear to impact other insects.

It remains unknown if insects other than ants access these baits. However, based on the intensive observations conducted during 2006 and 2007, no insects other than ants were observed near the bait particles, so it is expected that the impact, if it occurs, is minimal.

In relation to the impact of *S. invicta* on insect abundance and taxa diversity, it is not surprising to have found no impacts at that level. Perhaps the negative impacts of *S. invicta* on other arthropods (excluding ants) is not reflected on their abundance simply

because this species puts pressure at population levels that are not reflected in numbers but in biomass and size which were not considered in this investigation.

Although credible experimental work links *S. invicta* to effects on invertebrates, claims about the impact of this species are built mostly on anecdotal observations (Allen et al 2004, Tschinkel 2006). To cite some examples without experimental evidence, *S. invicta* has been linked to the declining horned lizard. *S. invicta* is believed to competitively exclude harvester ants, *Pogonomyrmex sp.*, one of the primary food sources of this lizard (Taber 1998). In another study, Barlett (1997) speculates that the decline of snakes in some areas is caused by egg predation by *S. invicta*. Another observational study argues that *S. invicta* exerts a significant impact on black capped vireo survival, a federally endangered species (Stake and Cimprich 2003). However, there are few studies involving manipulations of *S. invicta* densities to explore the effect of their removal on invertebrates. The lack of field manipulative experiments and limited inferences that can be drawn from these experiments are strong justification for studying the impact of this invasive species on other insect species. Understanding the effects of *S. invicta* (positive, negative, none) as well the mechanisms underlying these effects will help to provide a better understanding of the impact of *S. invicta* on food webs. For achieving this, long term comprehensive ecological studies with a larger scope, conducted with controls and adequate temporal and spatial replication to provide an understanding of the effects of *S. invicta* on vertebrate and invertebrate populations

This study raises the level of certainty that bait insecticides and native ants are compatible at some level. Long term studies and replication of this study in other

habitats and other states is needed to completely understand the potential of using baits to enhance interspecific competition by native ants. However I have shown that baits do not impact native ants when the latter are at low and medium densities. It remains unclear whether the effect of interspecific competition has a net effect on fitness of *S. invicta*. Ecological interactions can also change geographically (Thompson 2005). Also, this study has shown that baits tilted the balance favoring some native ant species. Conversely, I have shown that repeated applications may harm native ants and that in areas with especially high native ant densities, baits may target resident fauna harming important species, causing “bait failures” and justifying the repeated use of bait insecticides to achieve “good” *S. invicta* control.

#### *Management Recommendations and Final Remarks*

Protocols to assess initial densities of both *S. invicta* and native ants may be necessary to design and implement management strategies that include establishing “native ant” thresholds for treatments or re-treatments to reduce the impact on non-target species and improve the efficacy of bait products, unless product companies develop more specific products initial assessments remains necessary especially in rangeland ecosystems.

These protocols may include the use of food lures for determining relative abundance and, more importantly, behavioral dominance on targeted areas. This study and a few others have shown that ant species recruiting to food lures will be also those likely to get insecticidal bait (King and Tschinkel, Calixto et al 2007b). Food lures have been suggested as one of the tools to evaluate relative abundance and dominance of *S. invicta*

and to trigger bait applications on pastures (Pereira 2003, Pereira and Porter 2005). Preliminary studies conducted in Texas (Calixto et al, in preparation) shown that food lures can provide a reliable estimation of relative abundance and dominance of *S. invicta*. Plots with low, medium and high *S. invicta* density were established and performance of food lures was evaluated. Recruitment in high density areas was higher and strongly correlated to colony density. In less than 20 minutes, more than 90% of the lures were saturated with *S. invicta* foragers with a similar response at medium densities. At low densities, lures were never saturated, leaving “unvisited” lures where other ant species were later observed.

Relying on these preliminary observations and the results herein presented, I suggest the use of at least ten food lure units (0.5 gm beef/chicken wieners), deployed every 20 m on an imaginary grid. Units should be left exposed for no more than 30 minutes, revisited and provided an estimated number of ants per unit. If more than 70% of lures are found saturated with *S. invicta*, one might argue it is “safe” to treat using insecticide. Under these conditions *S. invicta* is expected to control the bait allowing native ants to escape the treatment. If only 50-70% of lures are occupied, but no other ant species is observed, a bait treatment may be relatively safe. However there are other considerations regarding whether bait should be applied to control *S. invicta* (ground temperature, humidity). Less than 50% of lures occupied does not warrant a safe application of baits. First, at this point, the treatment would not be necessary, numbers may be below the economic threshold, if one exists, for that particular site. Second, a bait application may impact ant species that are present at the moment and successful *S. invicta* suppression

would probably require repeated applications thus increasing economic costs of *S. invicta* management.

Also, timing of bait applications is important, *S. invicta* and native ants are not always active (through the day and seasons). Several studies have shown that *S. invicta* is likely active through the year compared to other ant species (Vogt et al 2005, Gibbons and Simberloff 2005). This allows treatments in periods of low native ant activity, like in the early spring in Texas (March) and later fall (December) (Calixto 2004). Time of day is also an important factor. Vogt et al (2005) indicated that bait treatments at noon and evening were likely to impact native ant species since *S. invicta* appears to forage more in the morning, late afternoon and evening, avoiding the high temperatures on the ground (Tschinkel 2006). Other ant species, like *Dorymyrmex sp.* and *Forelius sp.* are more resistant to high temperatures and are observed increasing foraging during warmer hours of the day. Other considerations to improve management practices are not here included such as the dietary requirements of *S. invicta* at specific times of the day or season. This information is necessary not only to develop better bait insecticide carriers but to avoid the amount of interspecific competition for these baits during the application.

All these recommended guidelines require more investigation. In order to validate these recommendations, investigation needs to focus on optimal foraging of *S. invicta* and competition for baits through day and season, using different matrices and performing applications on different localities with different ant assemblages, but where *S. invicta* is considered an economic pest. No data is yet available addressing issues such

as optimal foraging of *S. invicta* on insecticide baits both through the day and the seasons. In this study I have shown that morning applications during the spring and fall effectively suppresses *S. invicta* when other ant species relative abundance are low but that fall applications may target ant species other than *S. invicta* that increase following its reduction during the spring.

Finally, if an elevated percentage (~ 30%) of native ants is observed occupying food lures, the more likely it is that the bait treatment will fail in reducing *S. invicta* to those control levels expected by insecticide baits. More importantly, the reduction of the native ants may leave open niches that most likely will be occupied by *S. invicta* and accentuate the economic and ecologic impact. It is important to understand that a sustainable management of *S. invicta* is necessary not only to reduce the costs of management practices but also to reduce the impact on non target species that at the end of the spectrum, may hold the key for an effective management of *S. invicta* in the US.

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## APPENDIX 1

## FIGURES

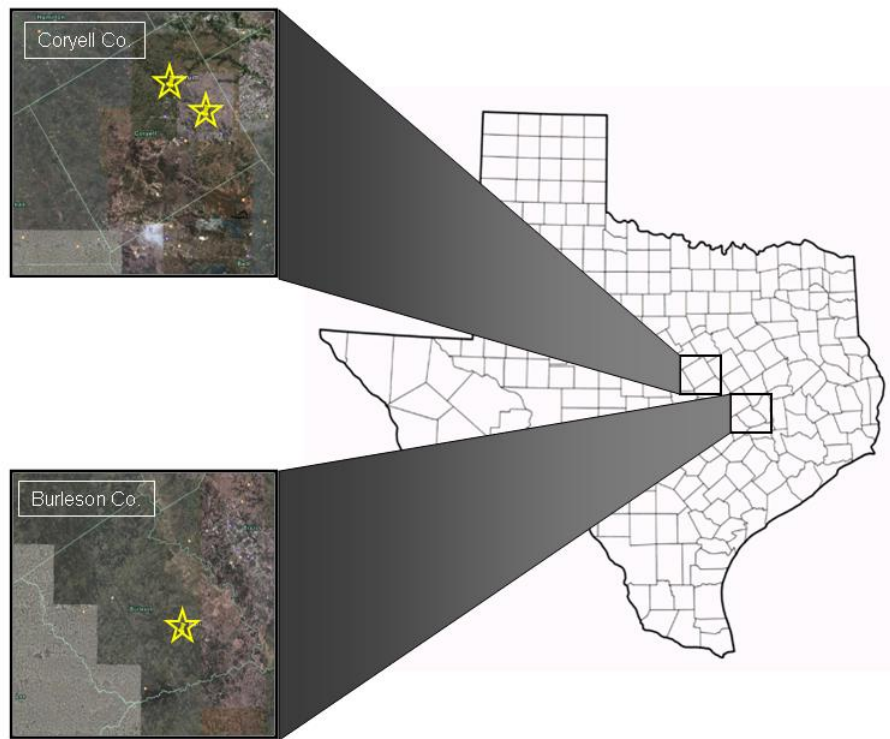


FIG. 1. Study sites located in Central Texas. Stars indicate the three areas where the investigation was conducted, two sites are located in Coryell Co. (“C3” and “Pruitt”) and the third site is located in Burleson Co. (“Barr”).

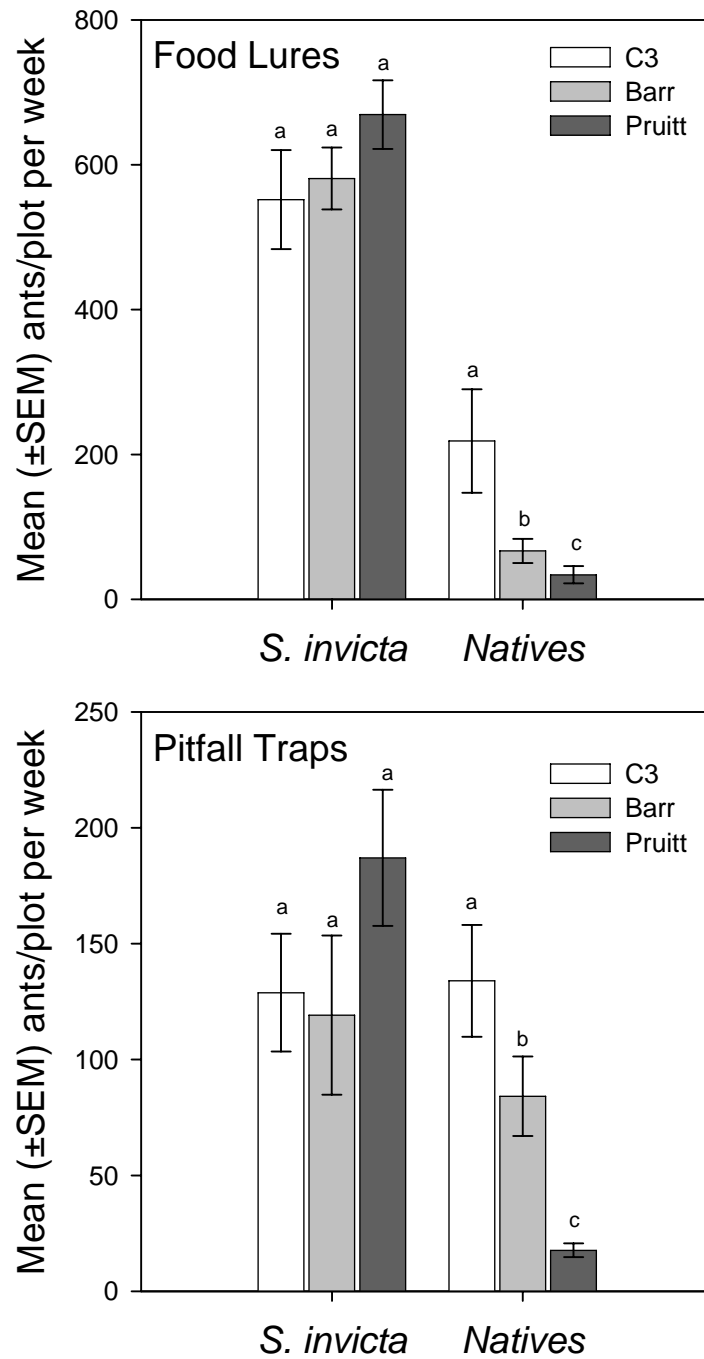


FIG. 2. Initial relative abundance and behavioral dominance (mean and  $\pm 1$  SE) for *S. invicta* and native ants based on food lures and pitfall traps sampled for seven weeks prior bait application; different letters signify  $P < 0.05$  (Linear Mixed Model).

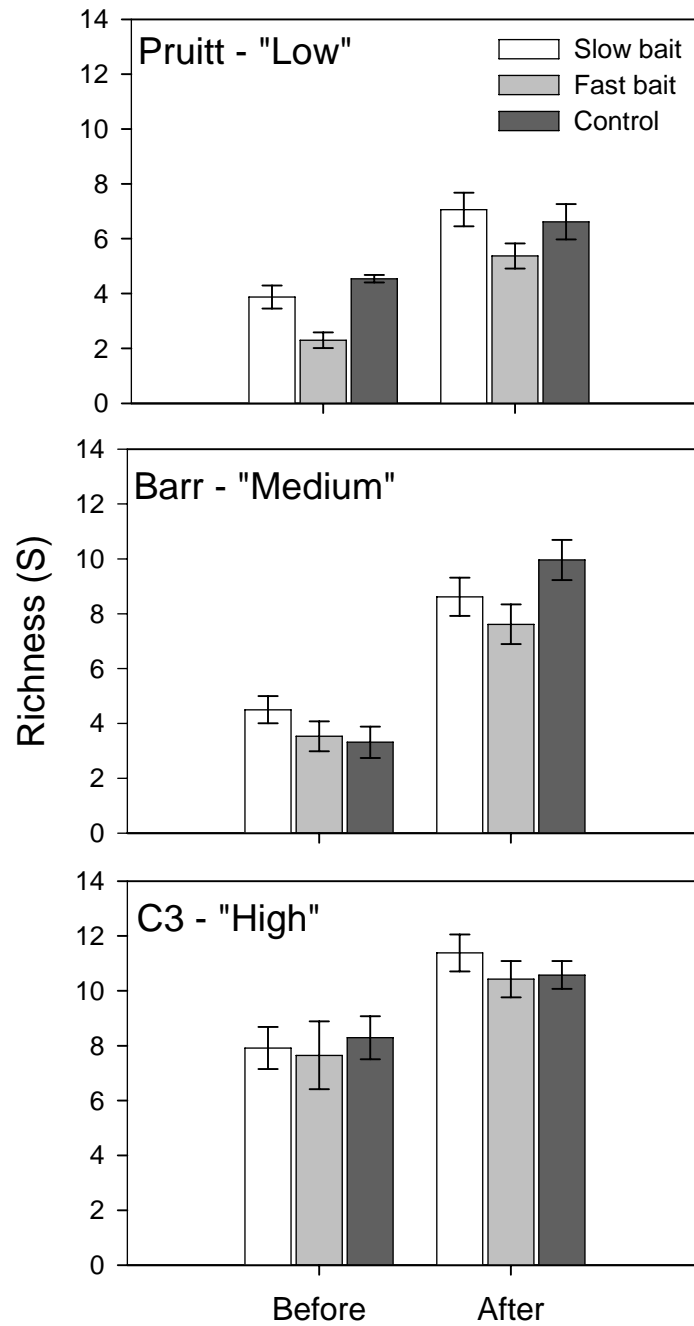


FIG. 3. Impact of baits and *S. invicta* reduction on species Richness ( $S$ ) based on pitfall traps, FY 2006.

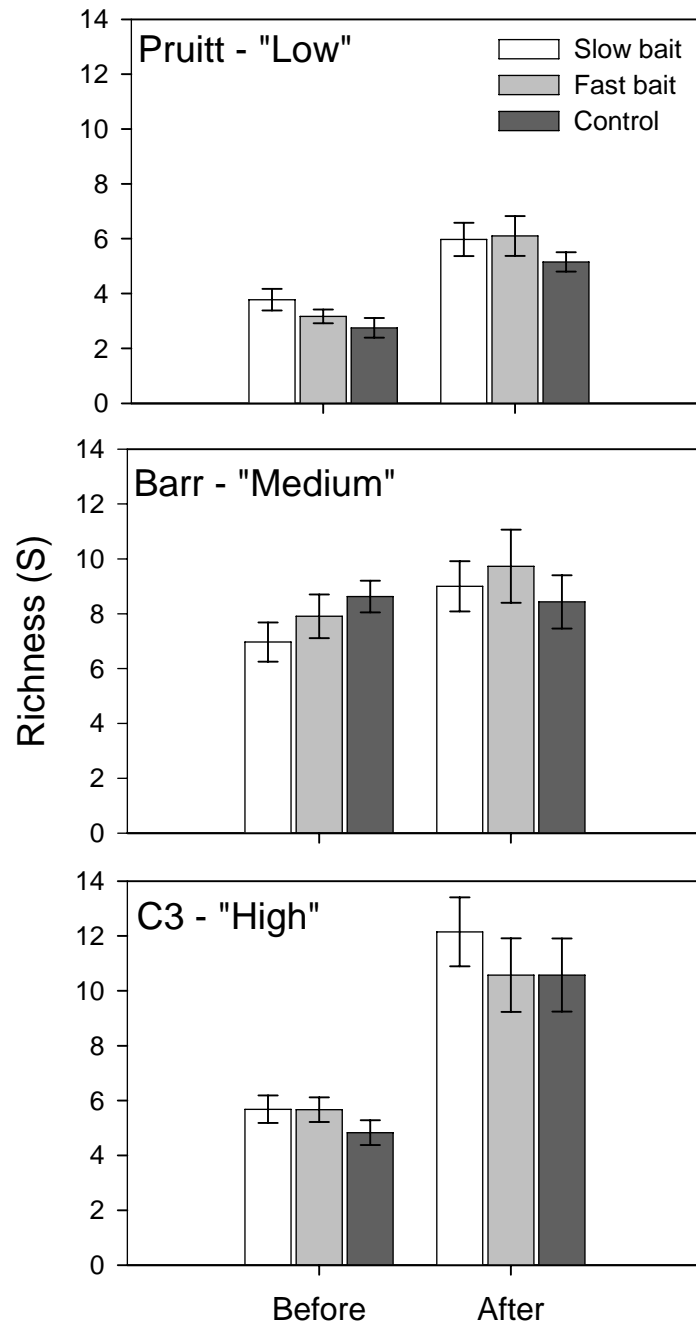


FIG. 4. Impact of baits and *S. invicta* reduction on species Richness (*S*) based on pitfall traps, FY 2007.

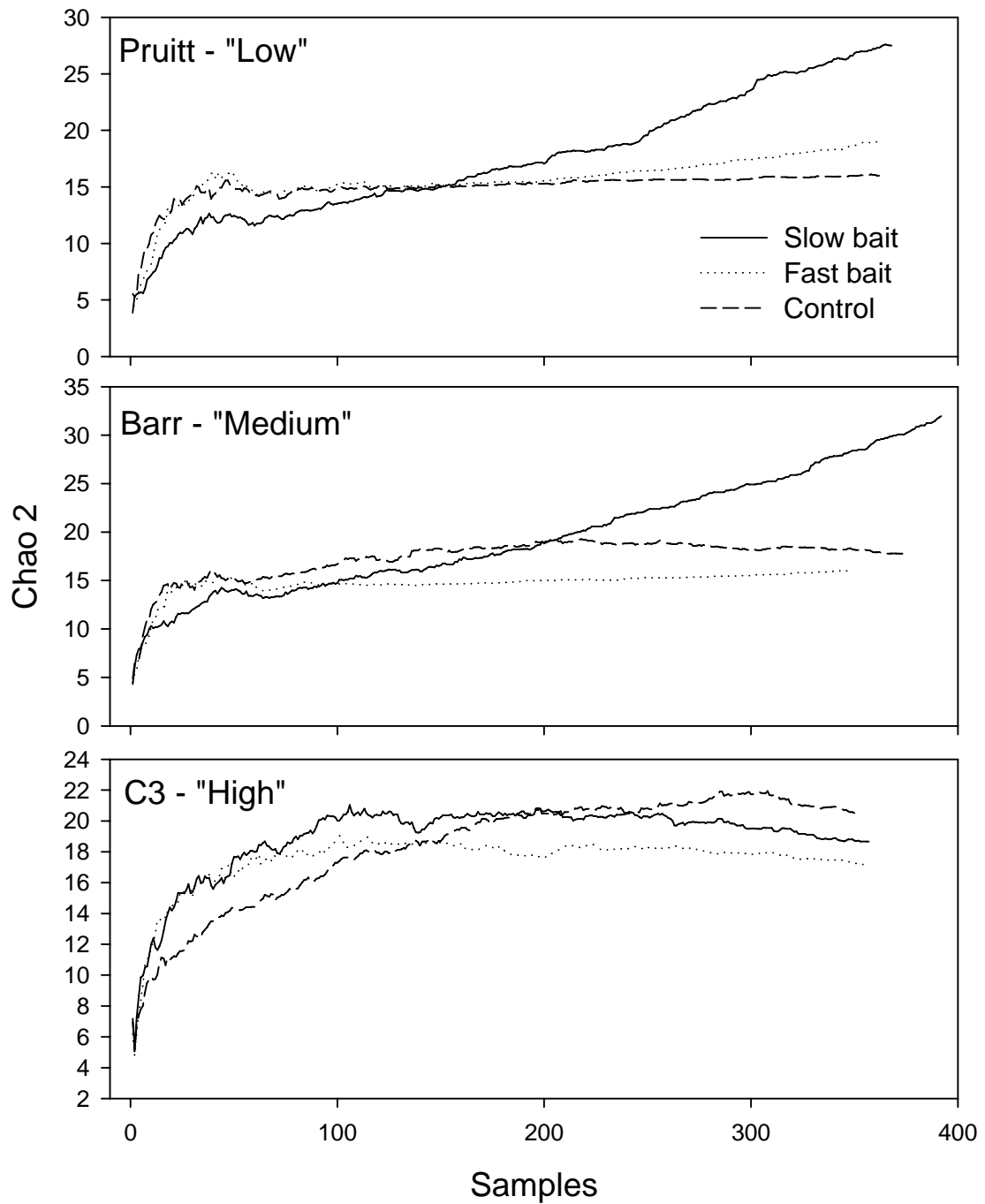


FIG. 5. Diversity estimators among different treatments (Chao2), FY 2006. Curves are sample based rarefaction curves generated from 50 randomizations of sample order.

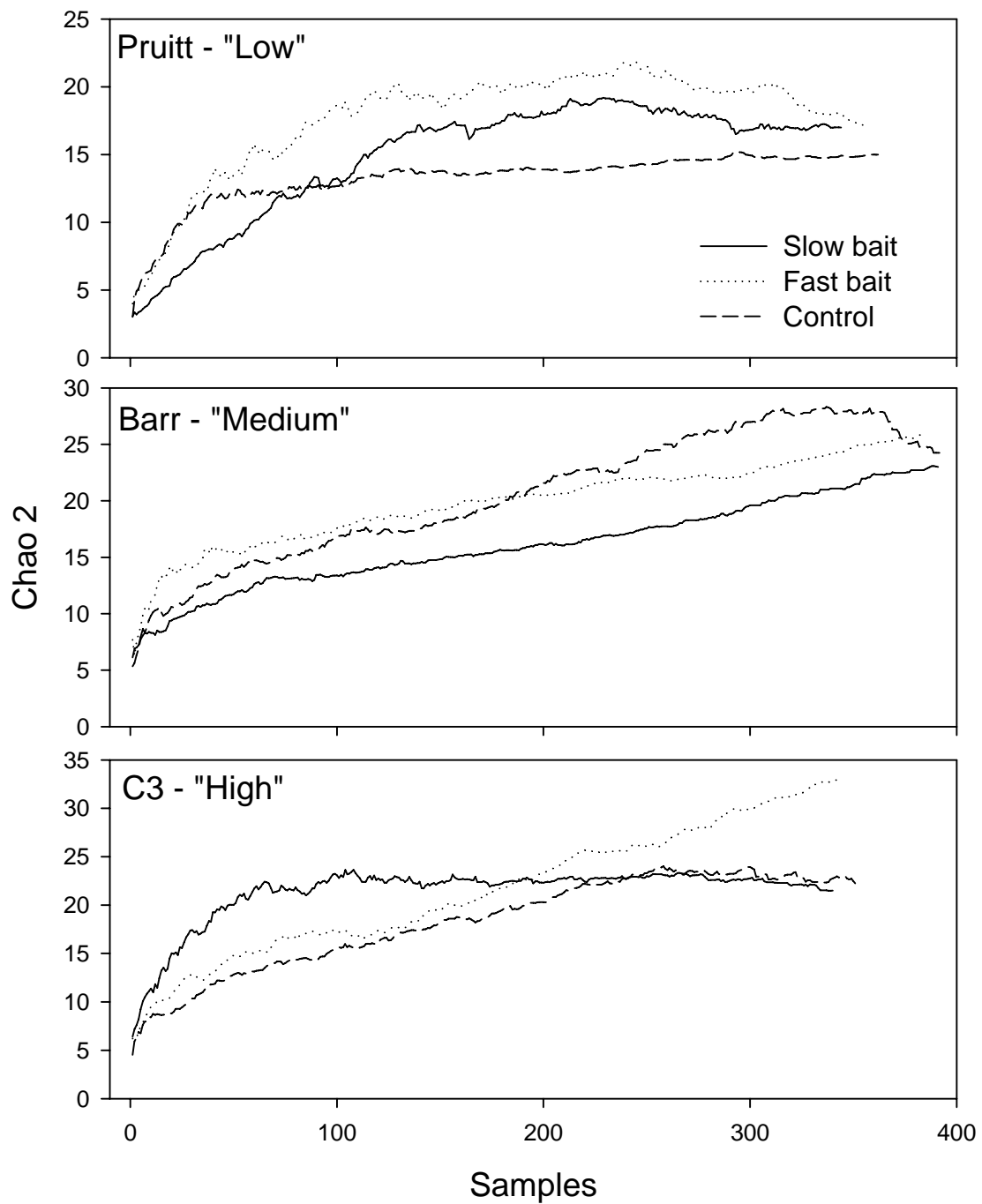


FIG. 6. Diversity estimators among different treatments (Chao2), FY 2007. Curves are sample based rarefaction curves generated from 50 randomizations of sample order.

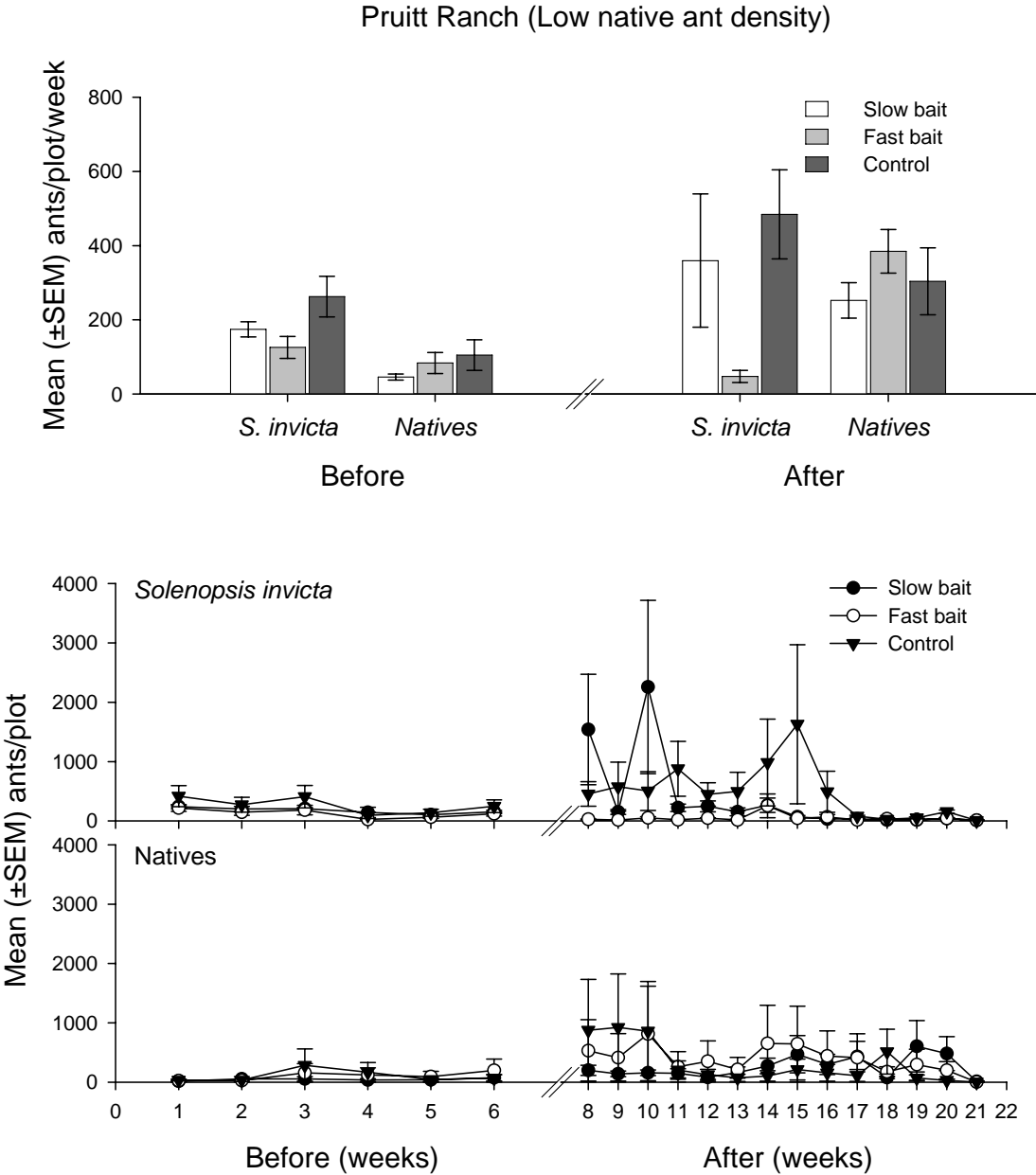


FIG. 7. Impact of baits on *S. invicta* and Natives based on pitfall traps, FY 2006.



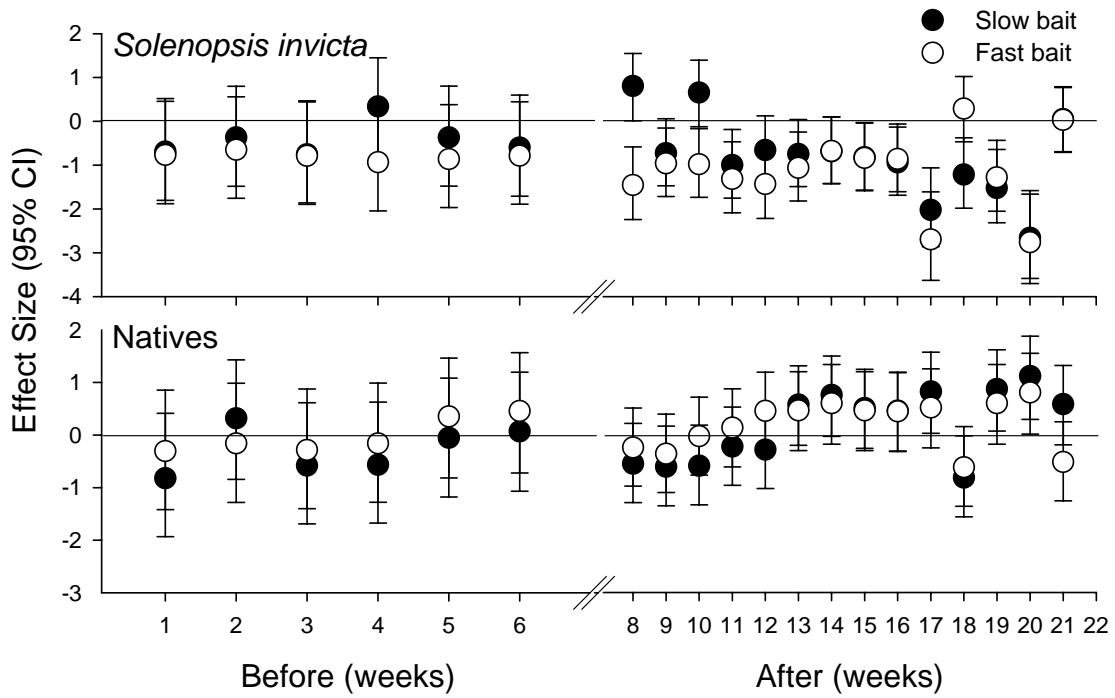


FIG. 8. Effect Sizes and 95% CI showing changes in pitfall traps in two treatments using BACI-P - Pruitt Ranch (Low Native Ant density), FY 2006.

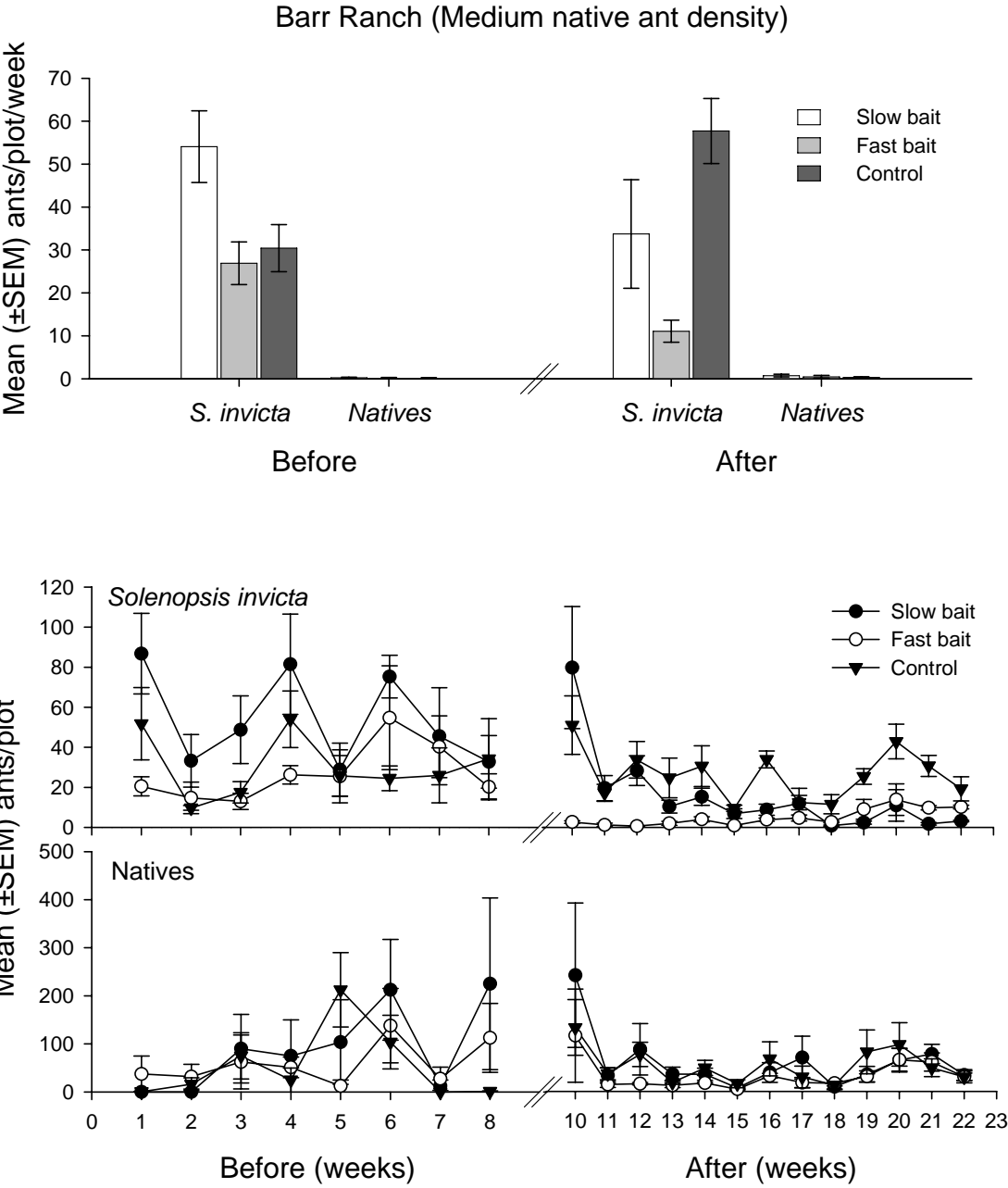


FIG. 9. Impact of baits on *S. invicta* and Natives based on pitfall traps, FY 2006.

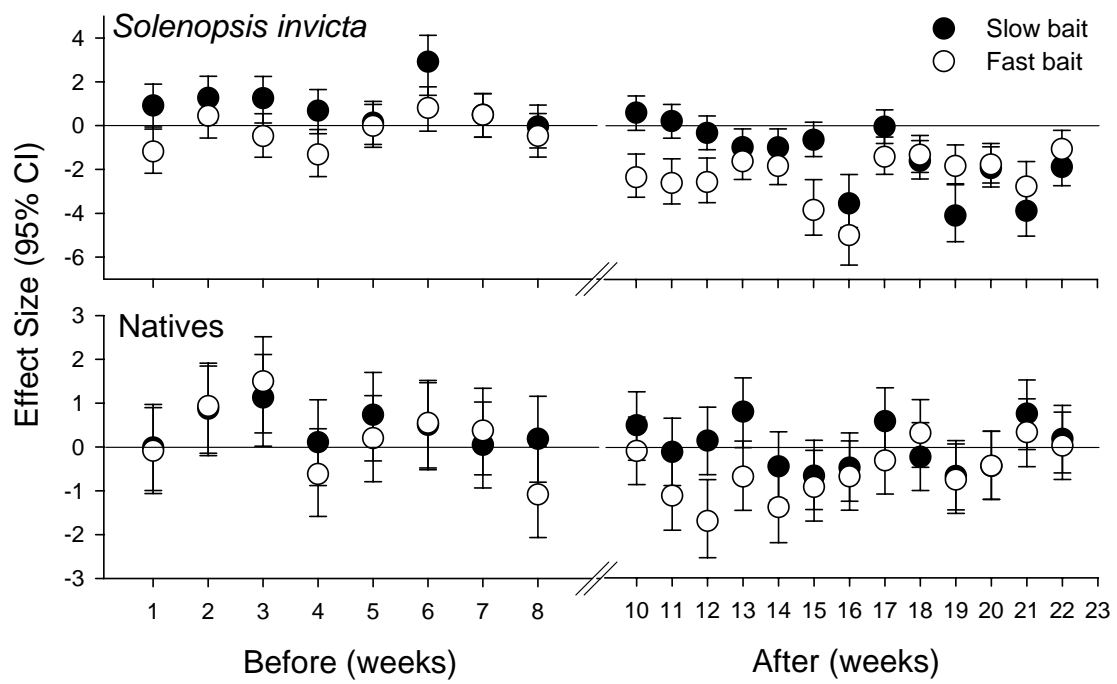


FIG. 10. Effect Sizes and 95% CI showing changes in pitfall traps in two treatments using BACI-P - Barr Ranch (Medium Native Ant density), FY 2006.

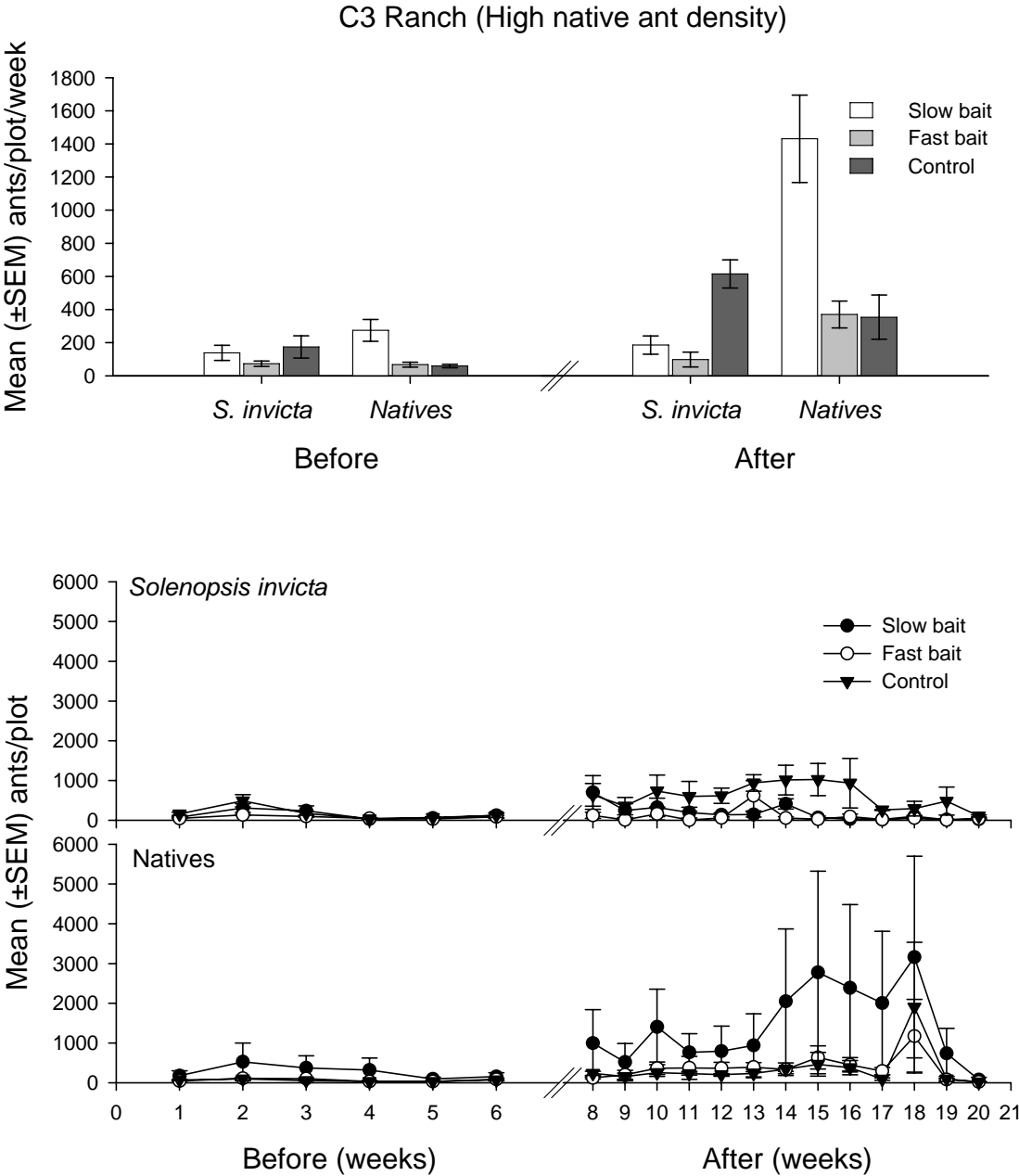


FIG. 11. Impact of baits on *S. invicta* and Natives based on pitfall traps, FY 2006.

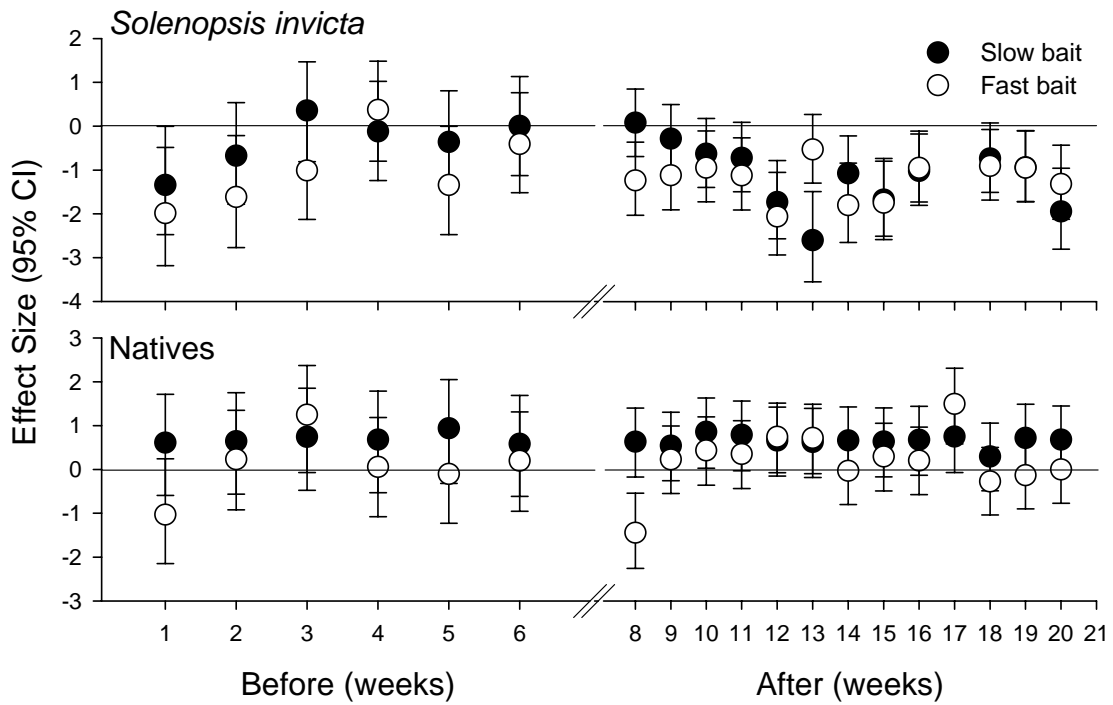


FIG. 12. Effect Sizes and 95% CI showing changes in pitfall traps in two treatments using BACI-P - C3 Ranch (High Native density), FY 2006.

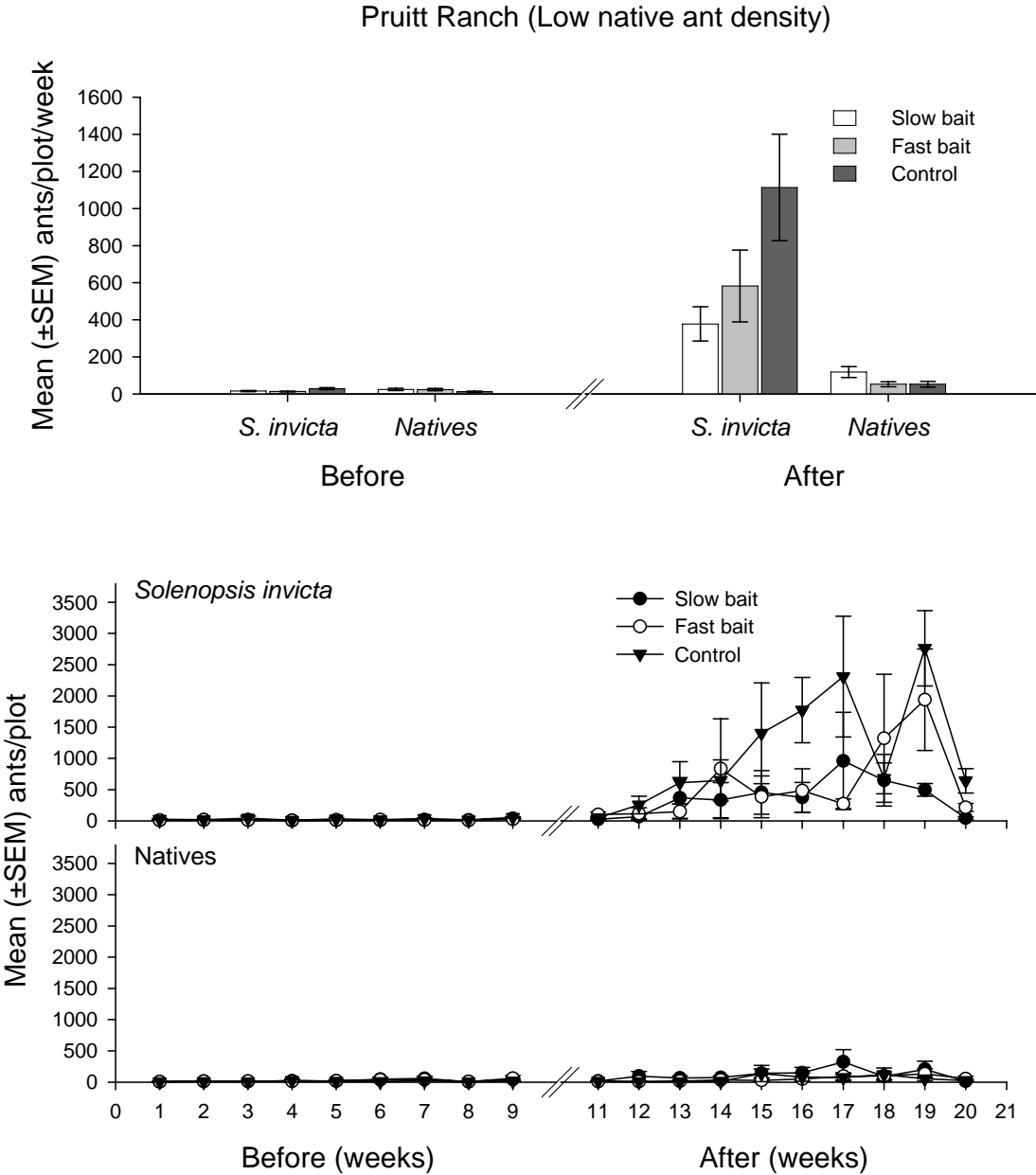


FIG. 13. Impact of baits on *S. invicta* and Natives based on pitfall traps, FY 2007.

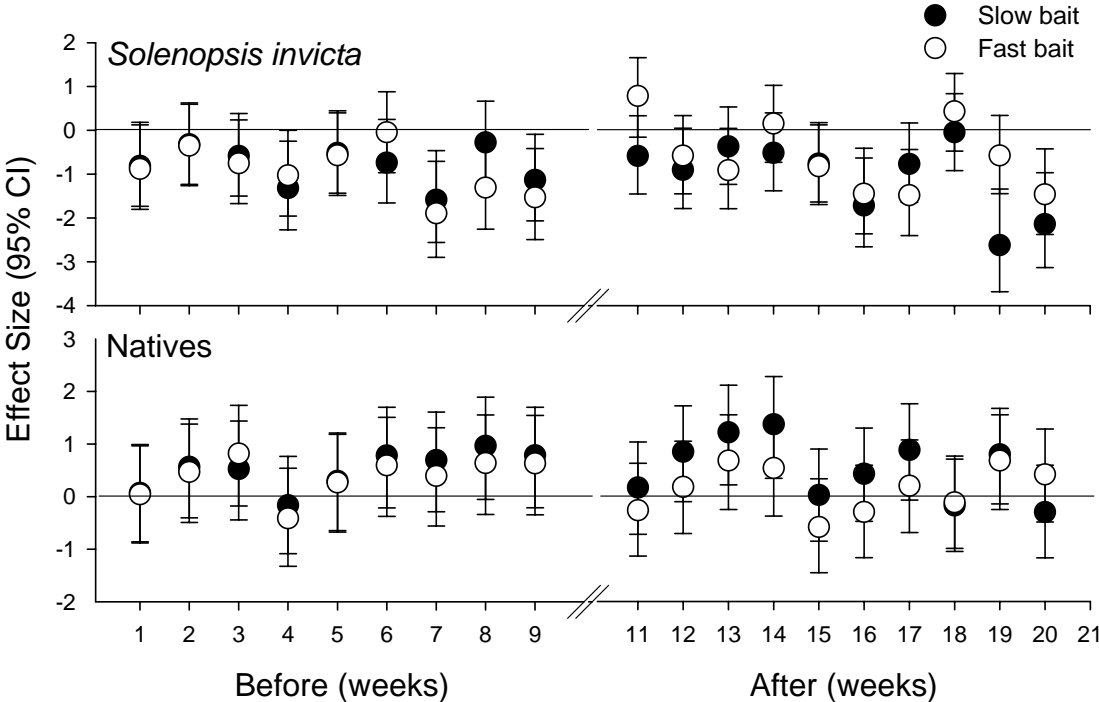


FIG. 14. Effect Sizes and 95% CI showing changes in pitfall traps in two treatments using BACI-P - Pruitt Ranch (Low Native ant density), FY 2007.

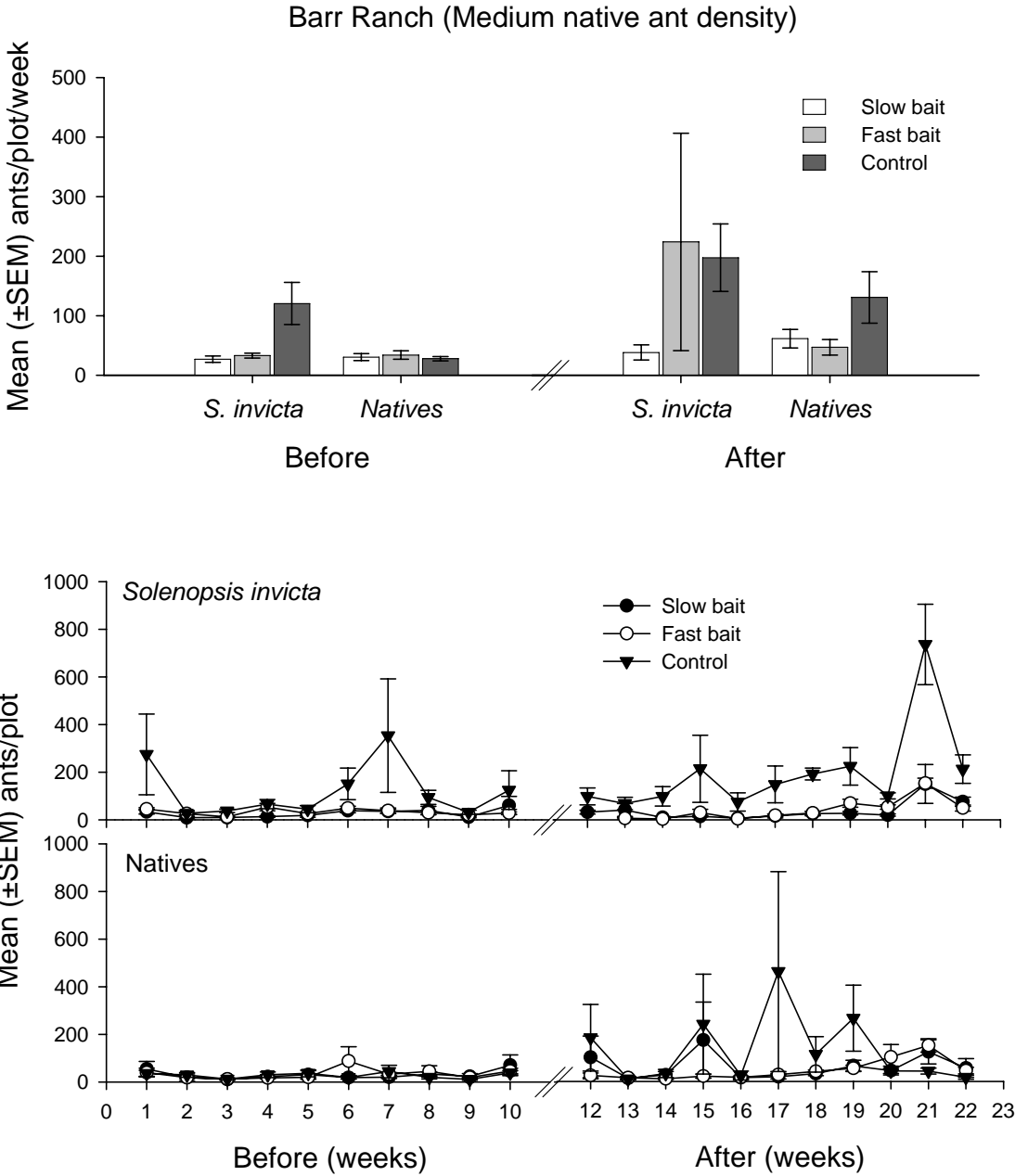


FIG. 15. Impact of baits on *S. invicta* and Natives based on pitfall traps, FY 2007.



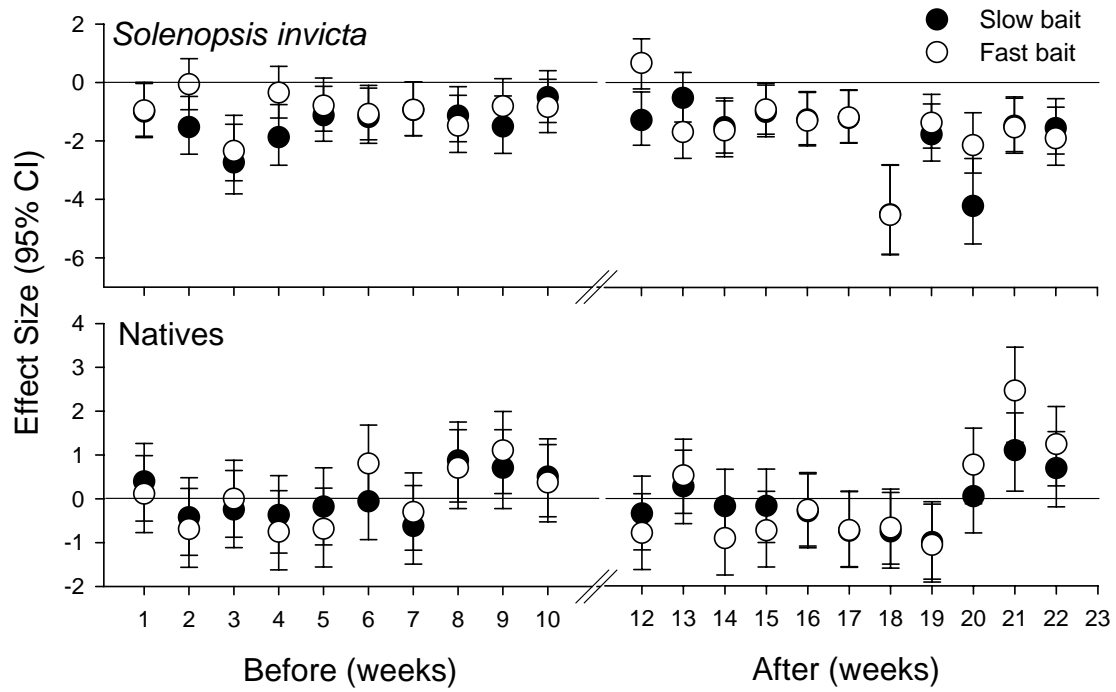


FIG. 16. Effect Sizes and 95% CI showing changes in pitfall traps in two treatments using BACI-P - Barr Ranch (Medium Native ant density), FY 2007.

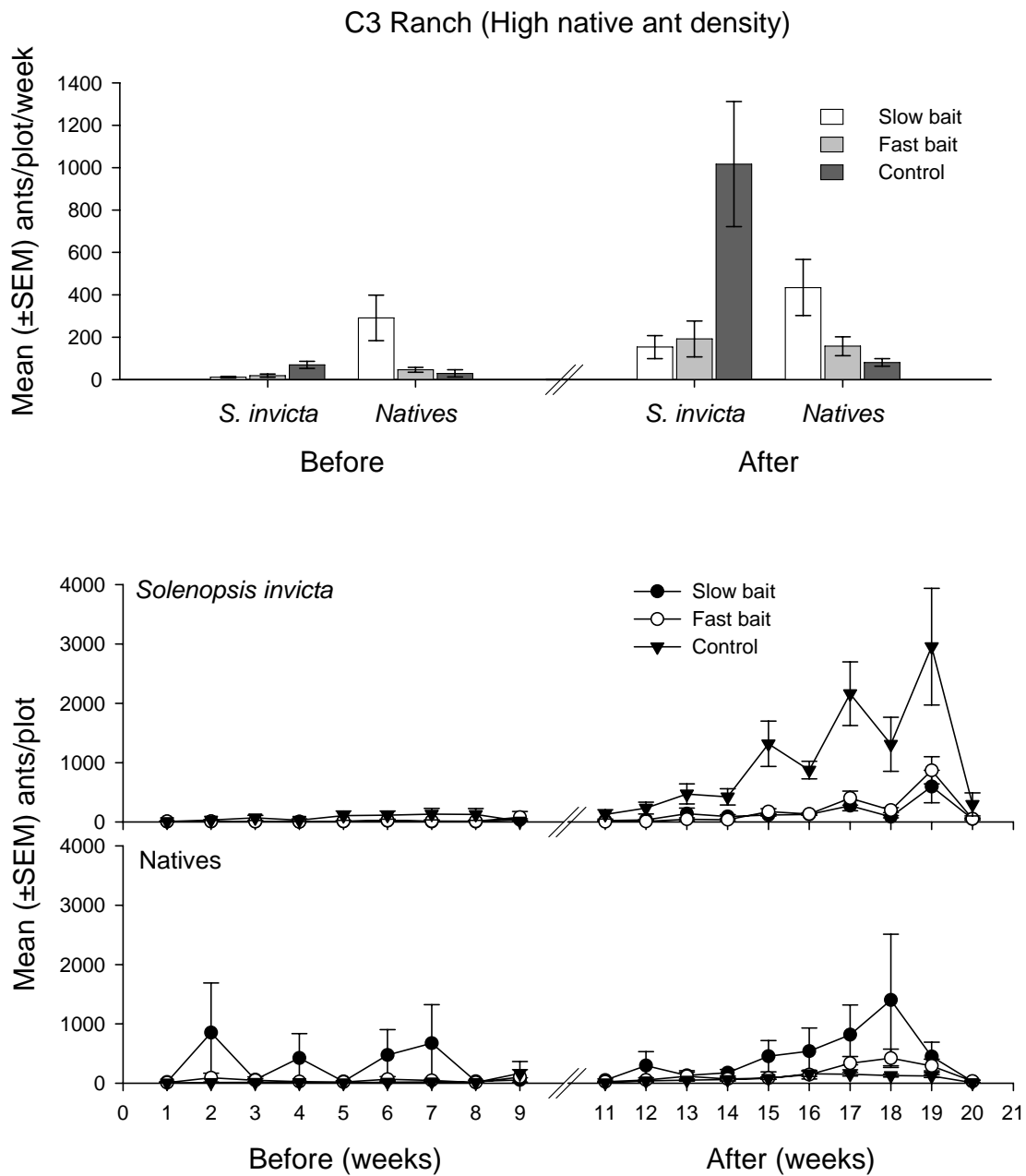


FIG. 17. Impact of baits on *S. invicta* and Natives based on pitfall traps, FY 2007.

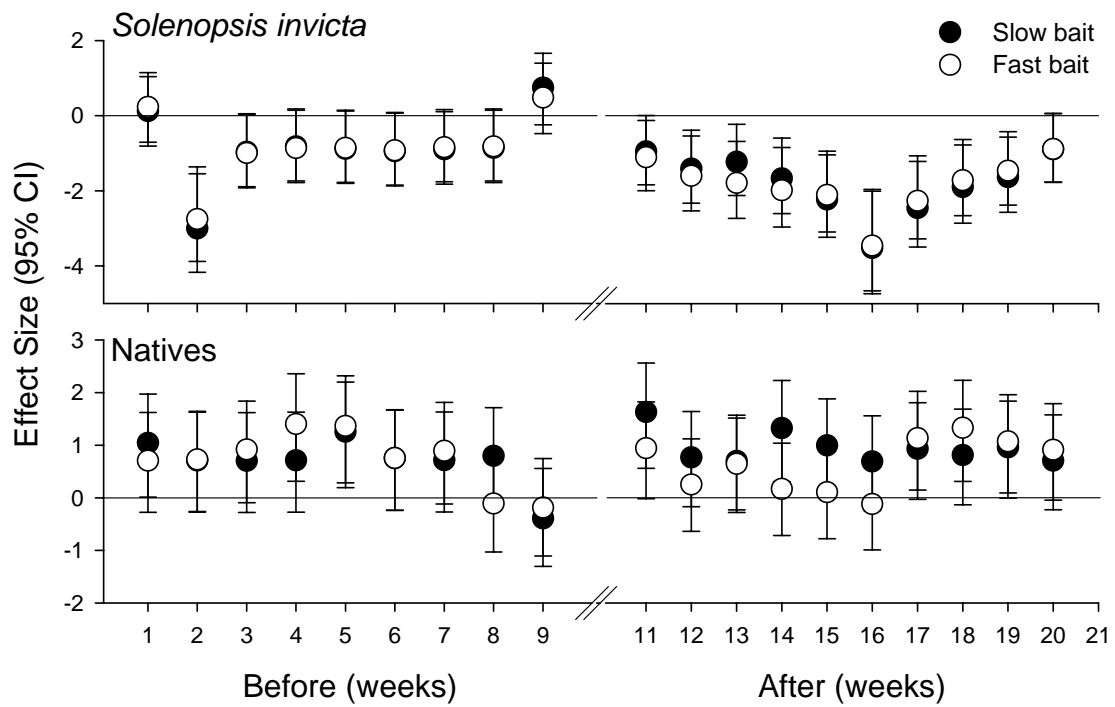


FIG. 18. Effect Sizes and 95% CI showing changes in pitfall traps in two treatments using BACI-P – C3 Ranch (High Native ant density), FY 2007.

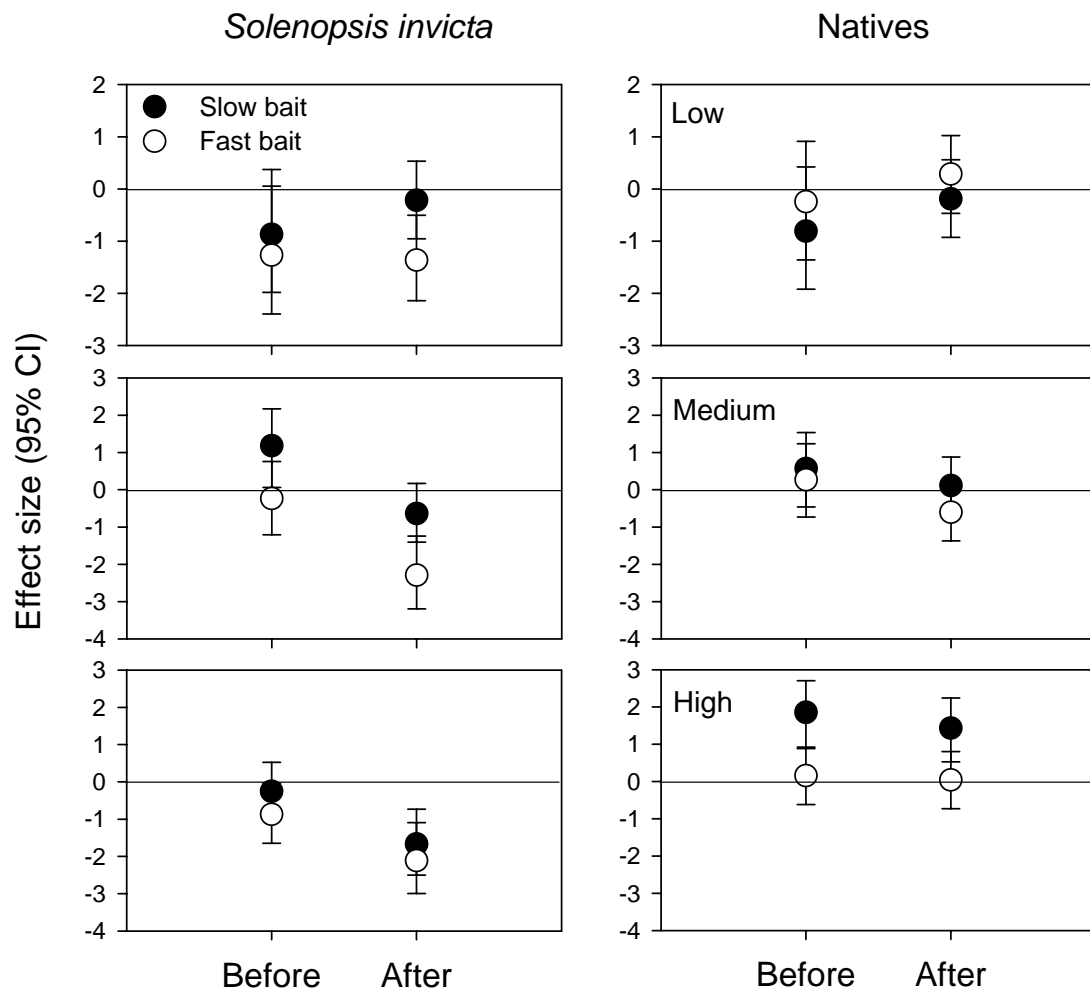


FIG. 19. Impact of baits on *S. invicta* and Natives (three densities, low, medium and high) based on pitfall traps, FY 2006.

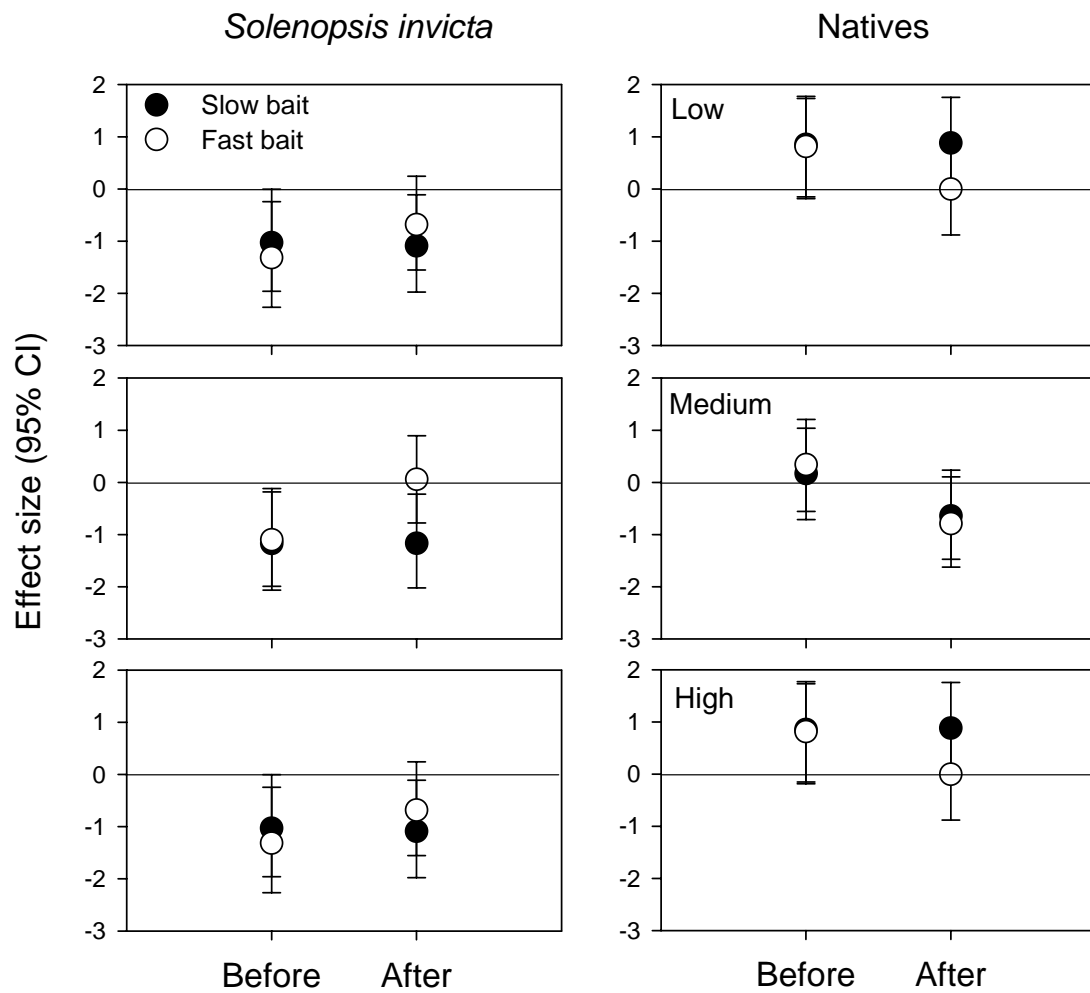


FIG. 20. Impact of baits on *S. invicta* and Natives (three densities, low, medium and high) based on pitfall traps, FY 2007.

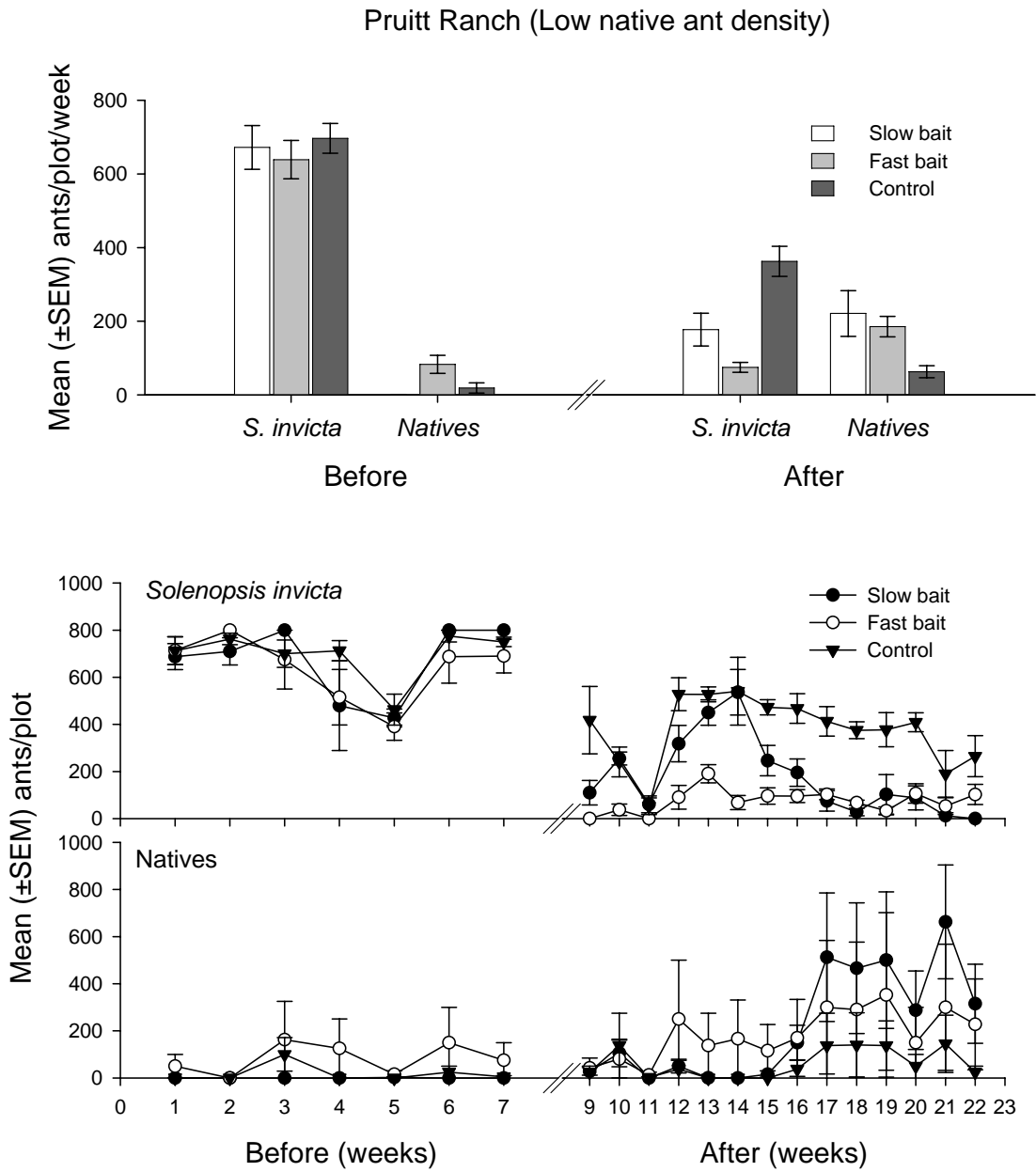


FIG. 21. Impact of baits on *S. invicta* and natives based on food lures, FY 2006.

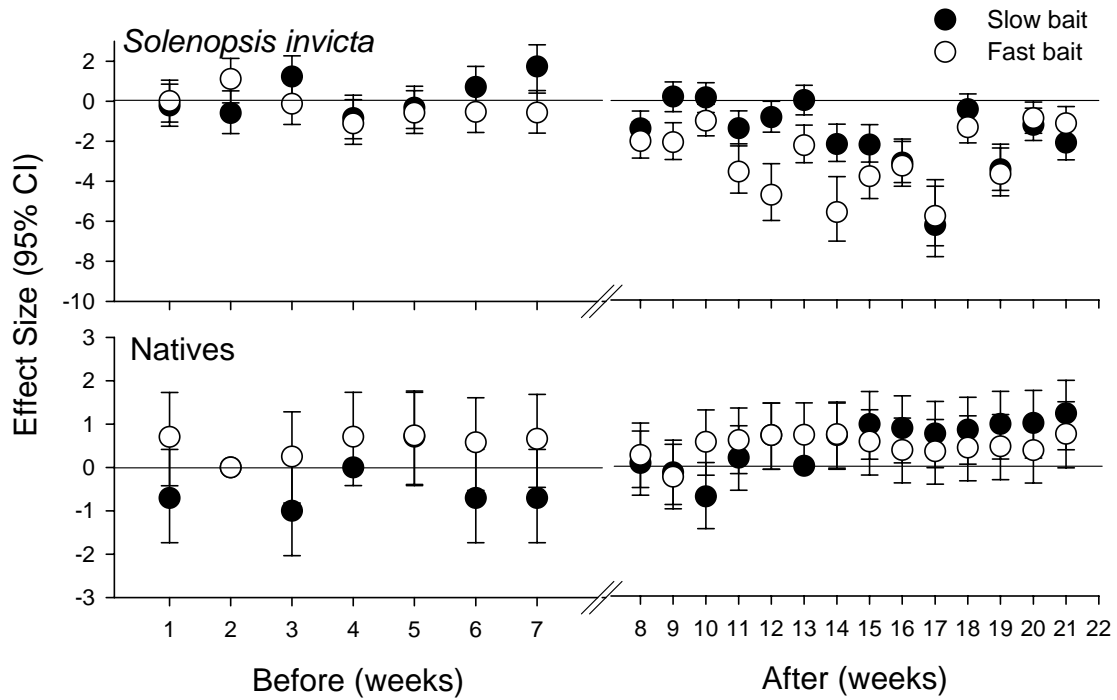


FIG. 22. Effect Sizes and 95% CI showing changes in food lures counts in two treatments using BACI-P - Pruitt Ranch (Low Native Ant density), FY 2006.

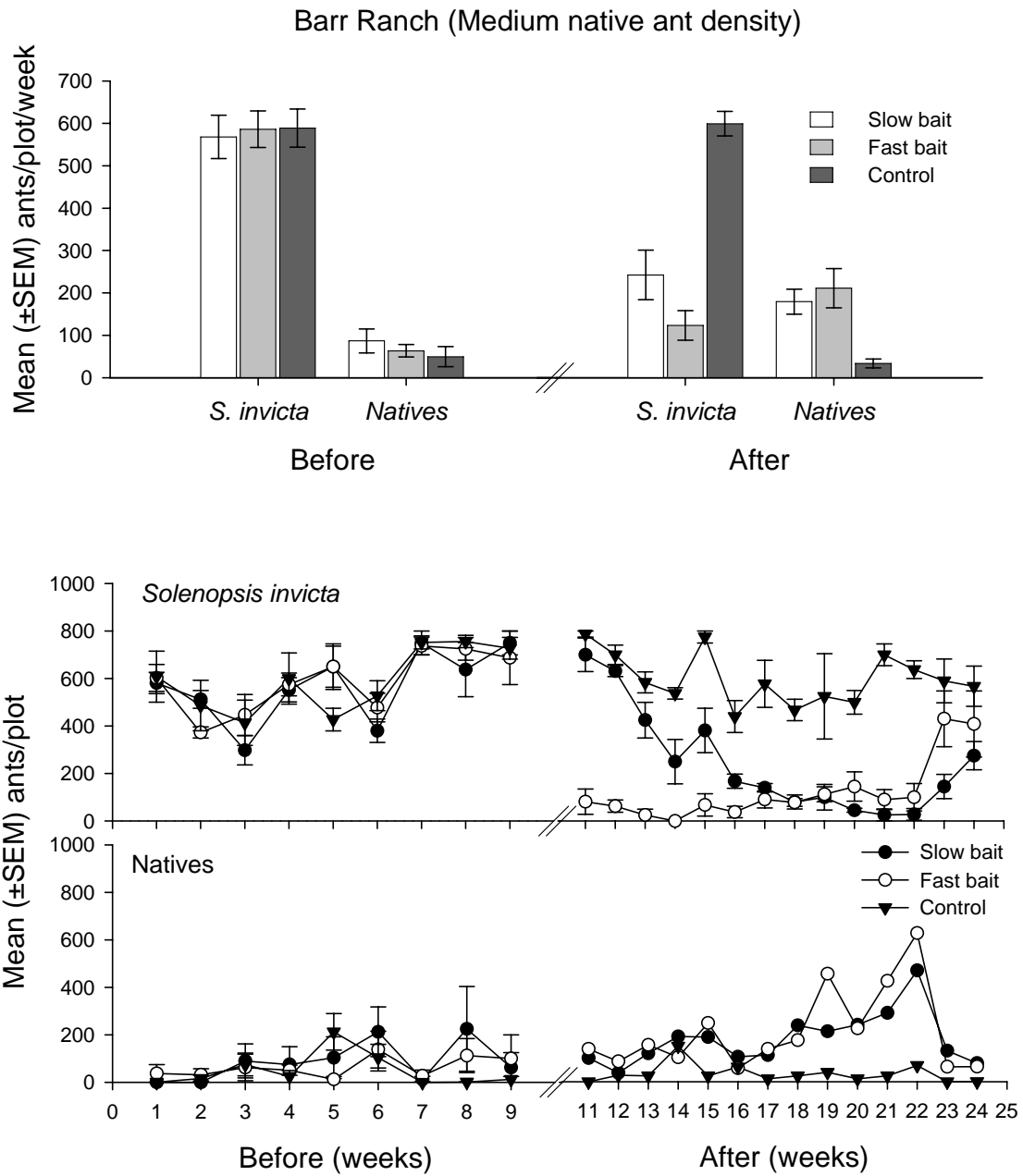


FIG. 23. Impact of baits on *S. invicta* and Natives based on food lures, FY 2006.



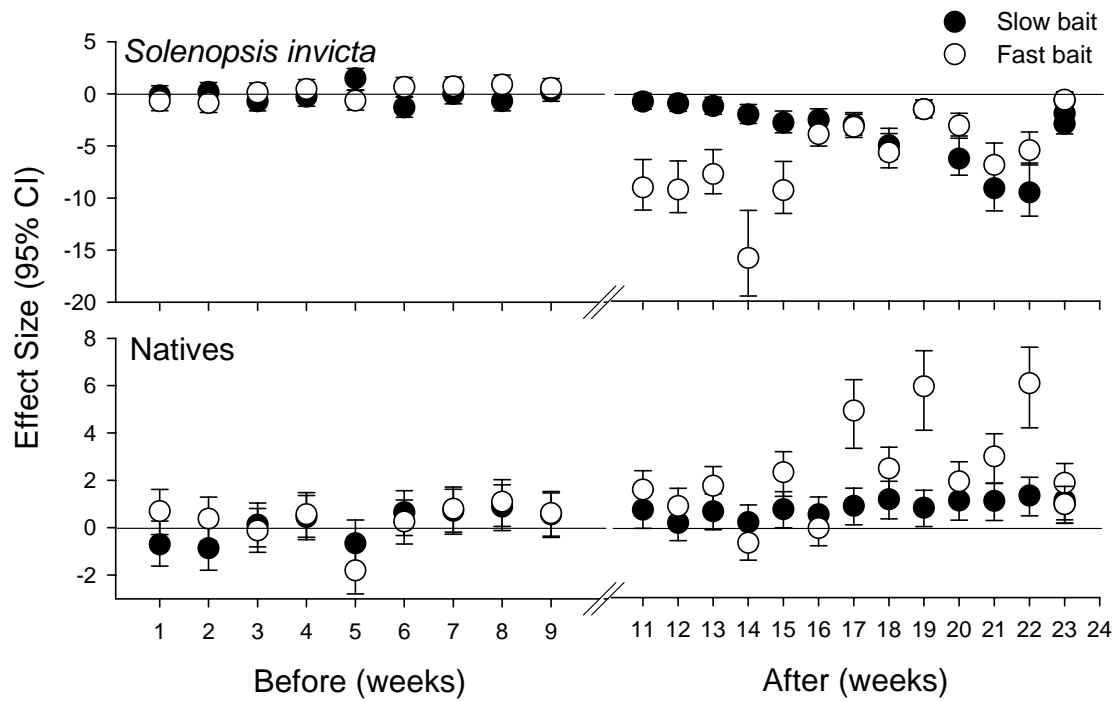


FIG. 24. Effect Sizes and 95% CI showing changes in food lures counts in two treatments using BACI-P - Barr Ranch (medium native ant density), FY 2006.

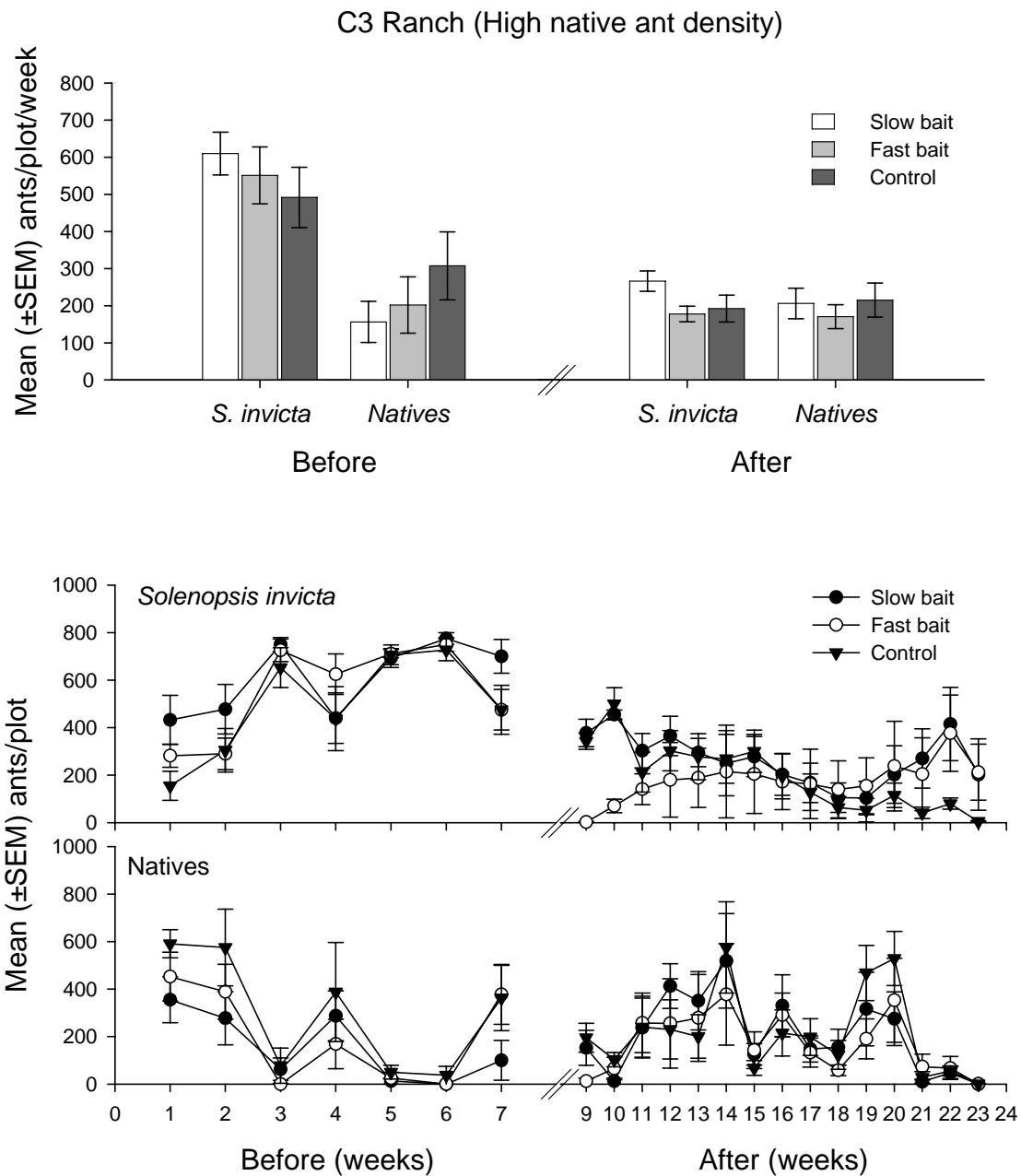


FIG. 25. Impact of baits on *S. invicta* and natives based on food lures, FY 2006.

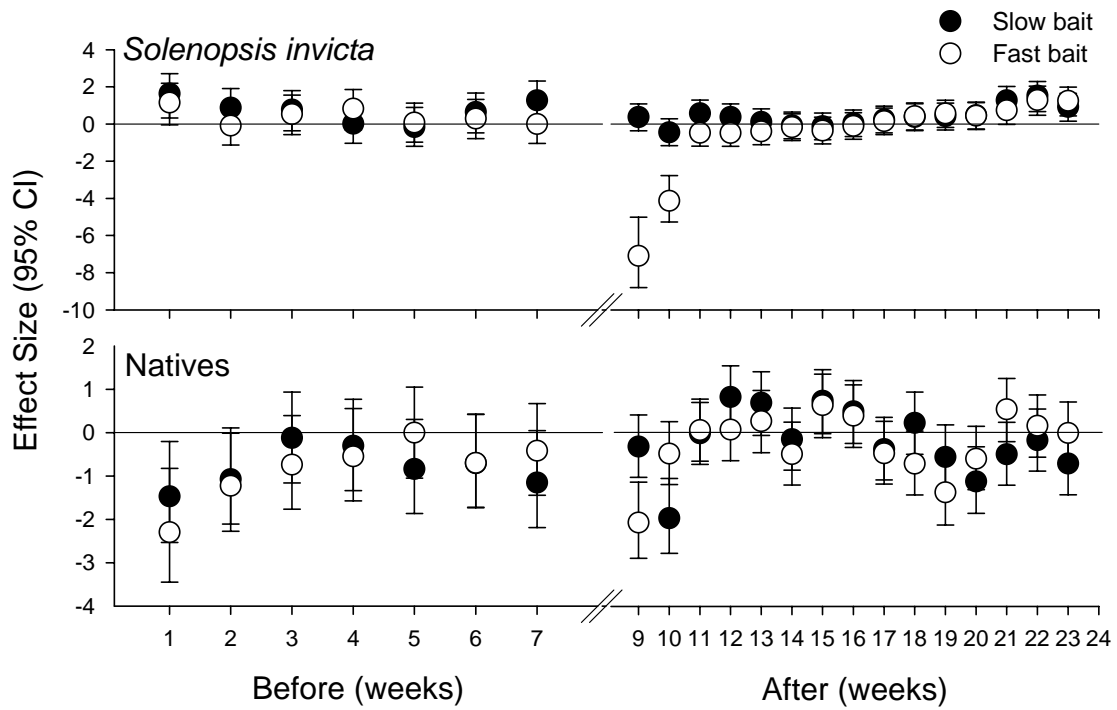


FIG. 26. Effect Sizes and 95% CI showing changes in food lures counts in two treatments using BACI-P - C3 Ranch (high native density), FY 2006.

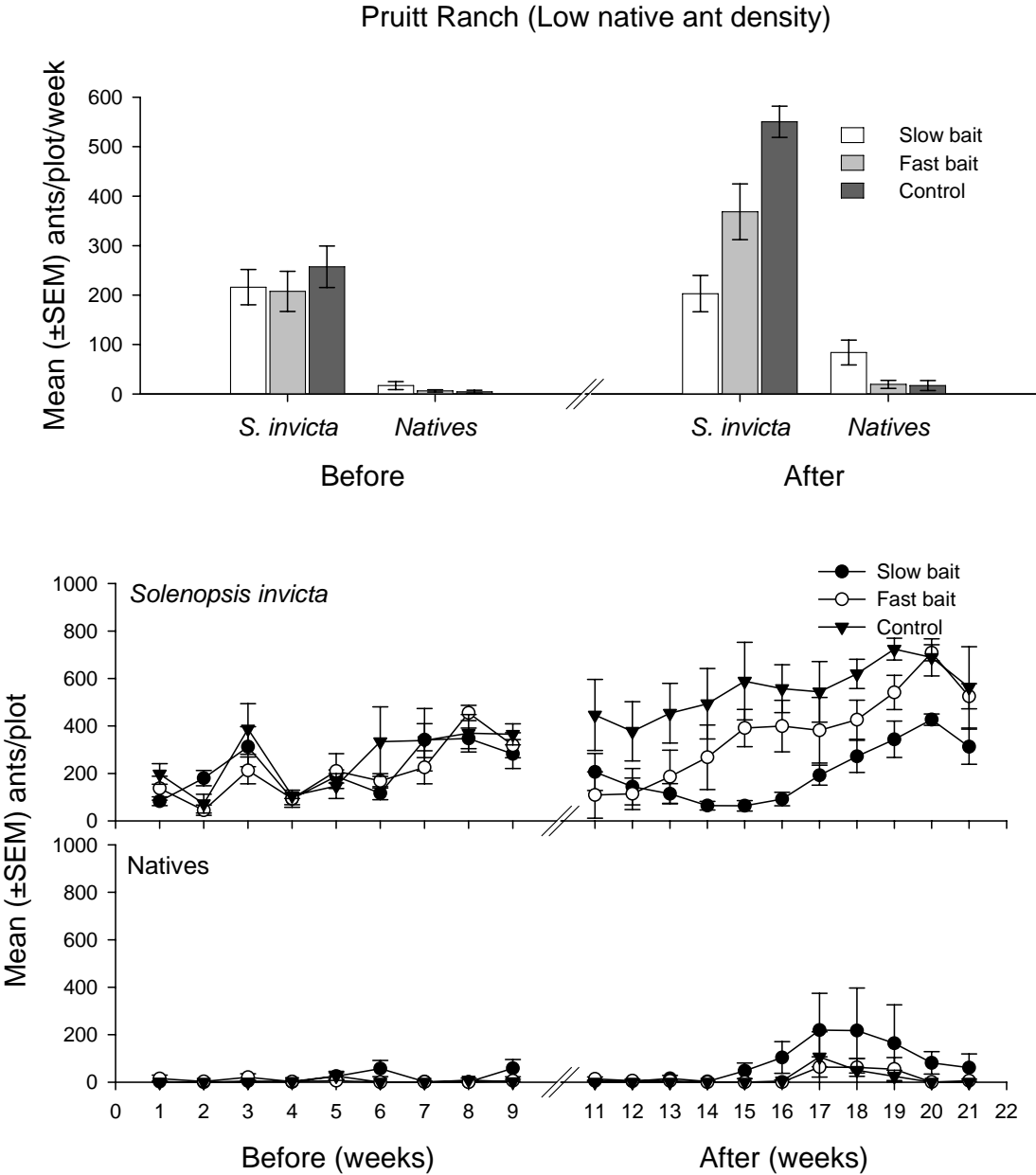


FIG. 27. Impact of baits on *S. invicta* and Natives based on food lures, FY 2007.

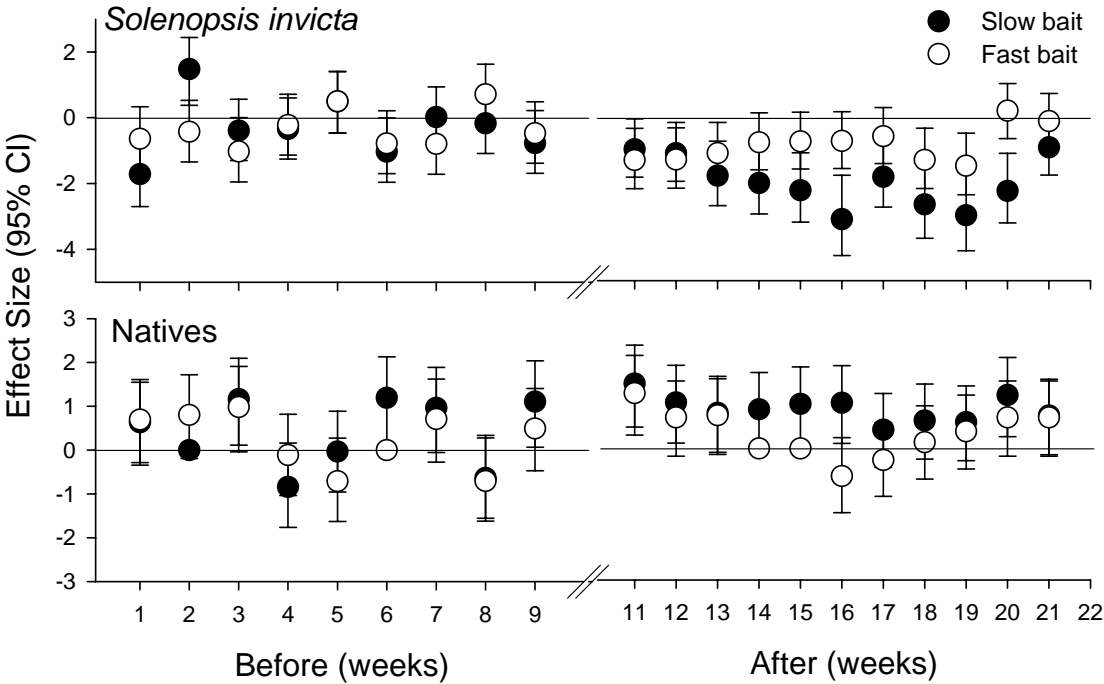


FIG. 28. Effect Sizes and 95% CI showing changes in food lures counts in two treatments using BACI-P - Pruitt Ranch (Low Native Ant density), FY 2007.

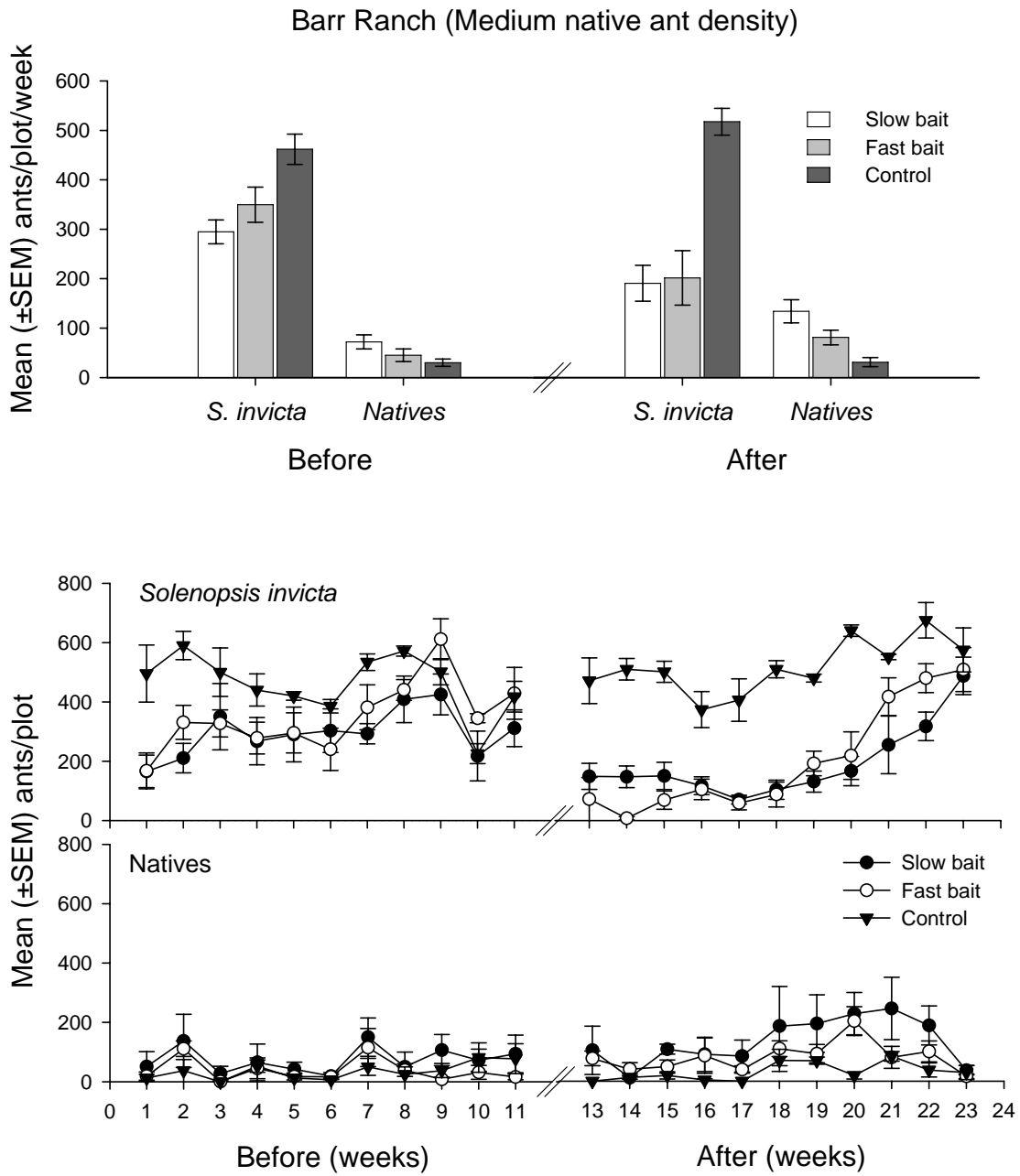


FIG. 29. Impact of baits on *S. invicta* and natives based on food lures, FY 2007.

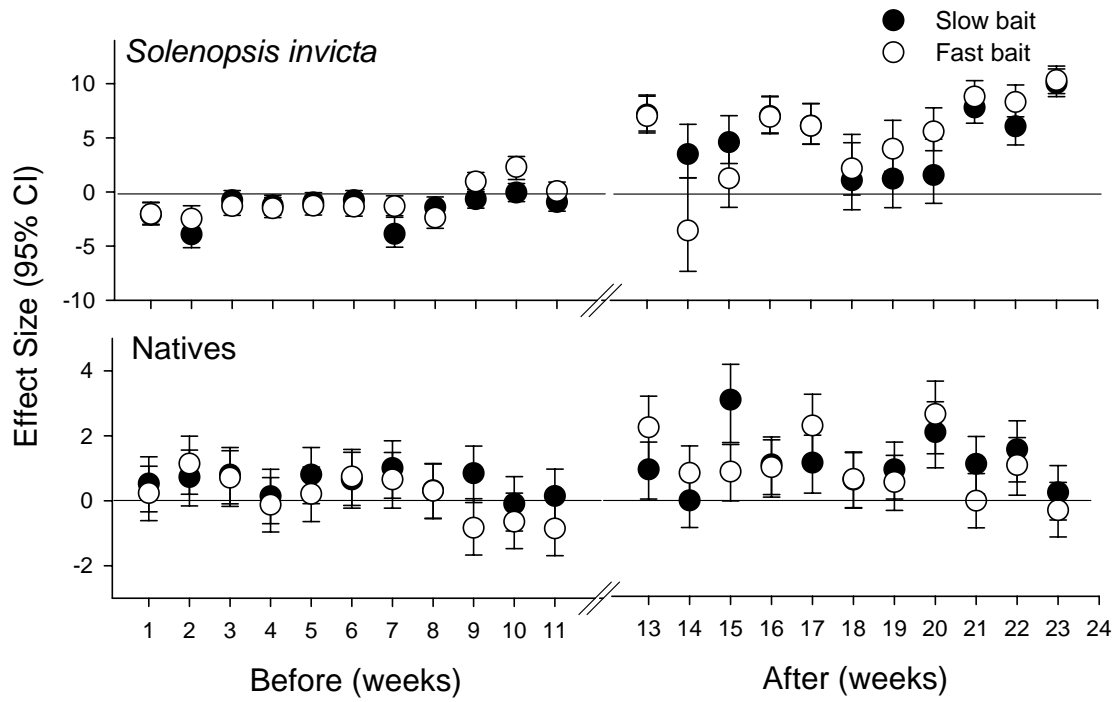


FIG. 30. Effect Sizes and 95% CI showing changes in food lures in two treatments using BACI-P - Barr Ranch (medium native ant density), FY 2007.

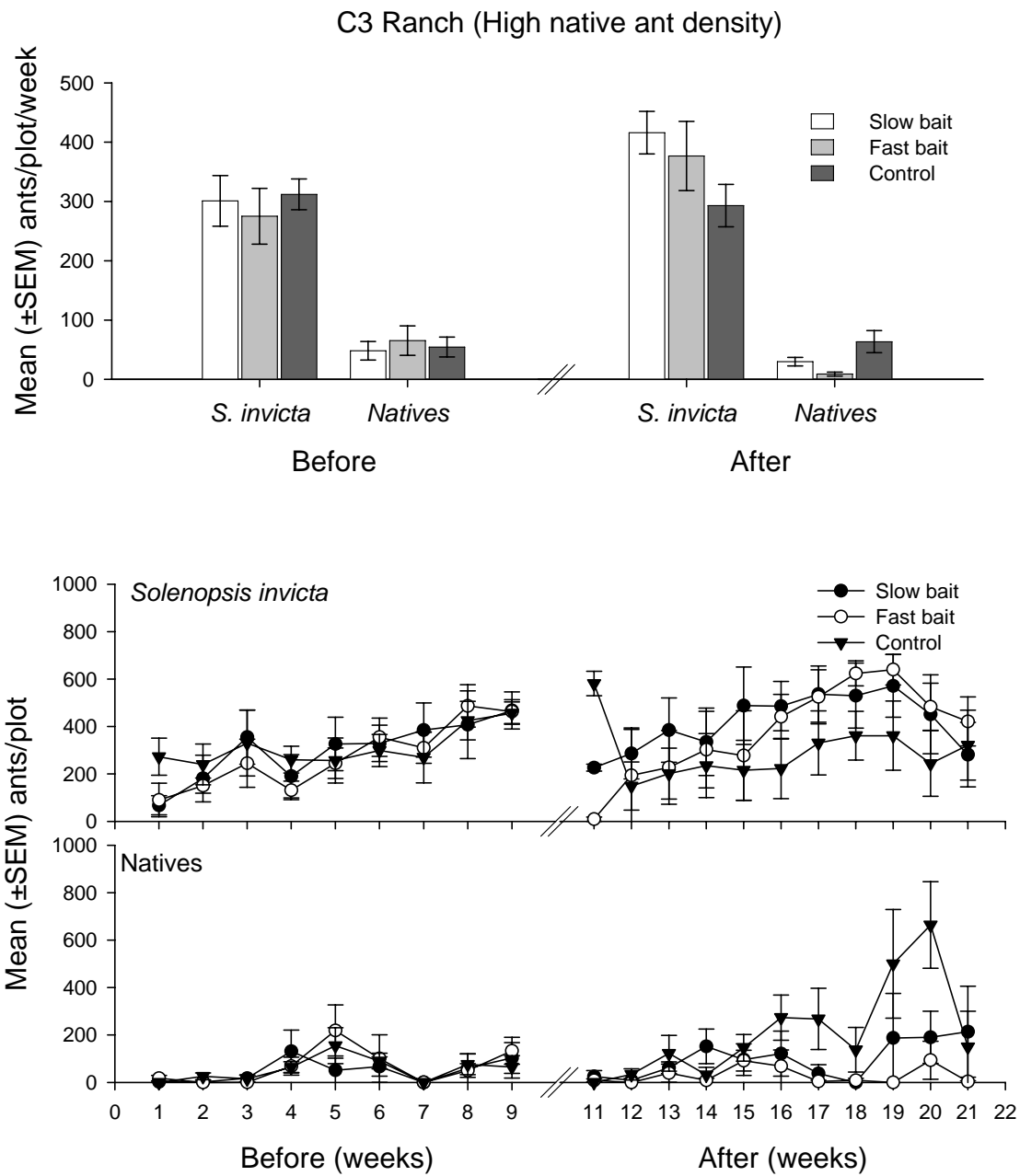


FIG. 31. Impact of baits on *S. invicta* and natives based on food lures, FY 2007.



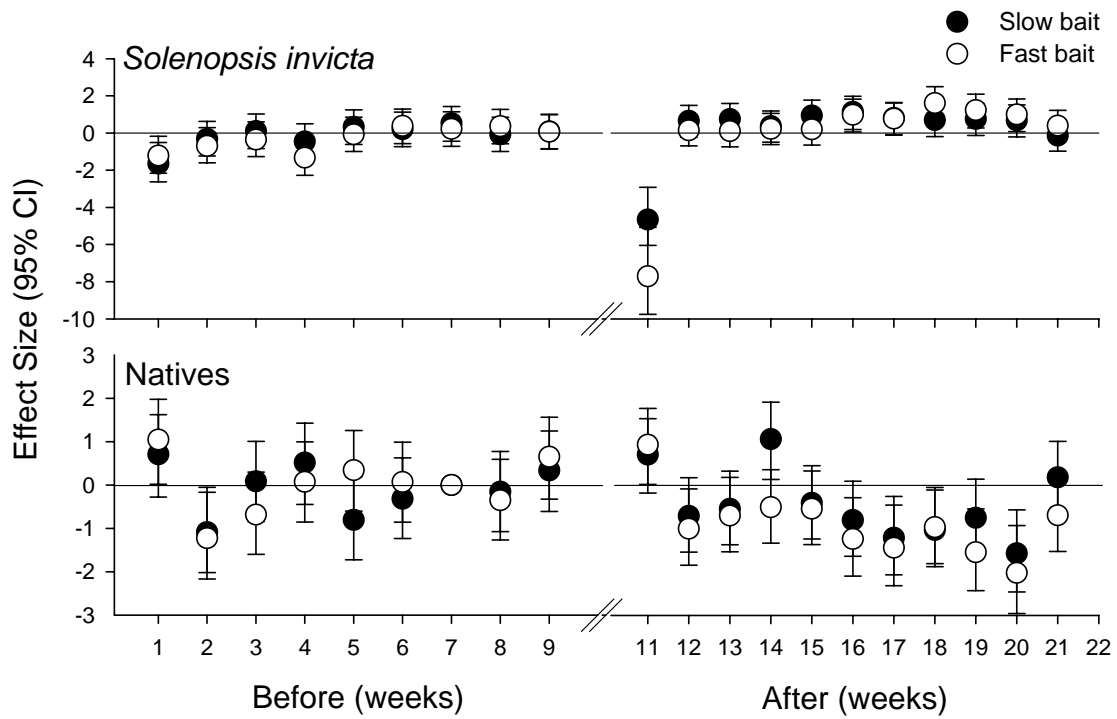


FIG. 32. Effect Sizes and 95% CI showing changes in food lures in two treatments using BACI-P – C3 Ranch (High Native Ant density), FY 2007.

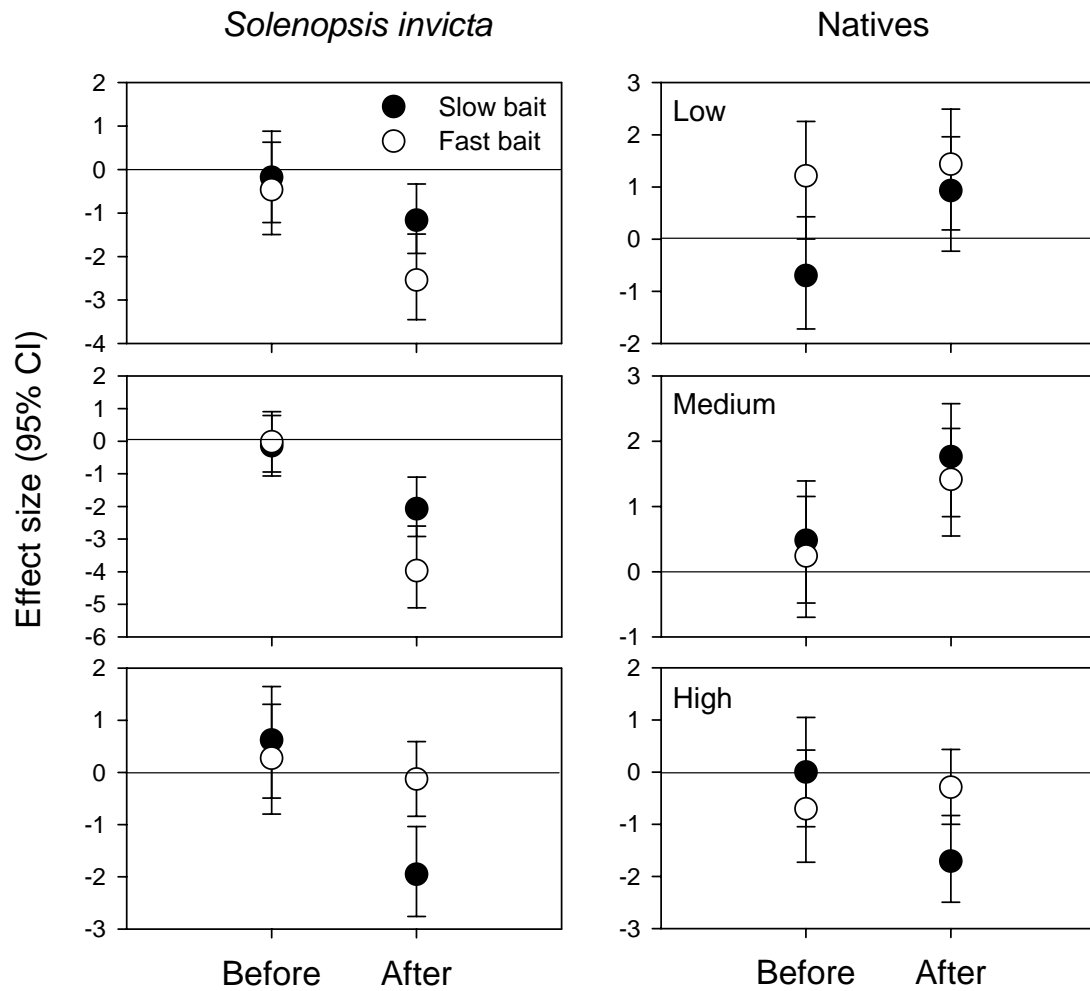


FIG. 33. Impact of baits on *S. invicta* and Natives (three densities, low, medium and high) based on food lures, FY 2006.

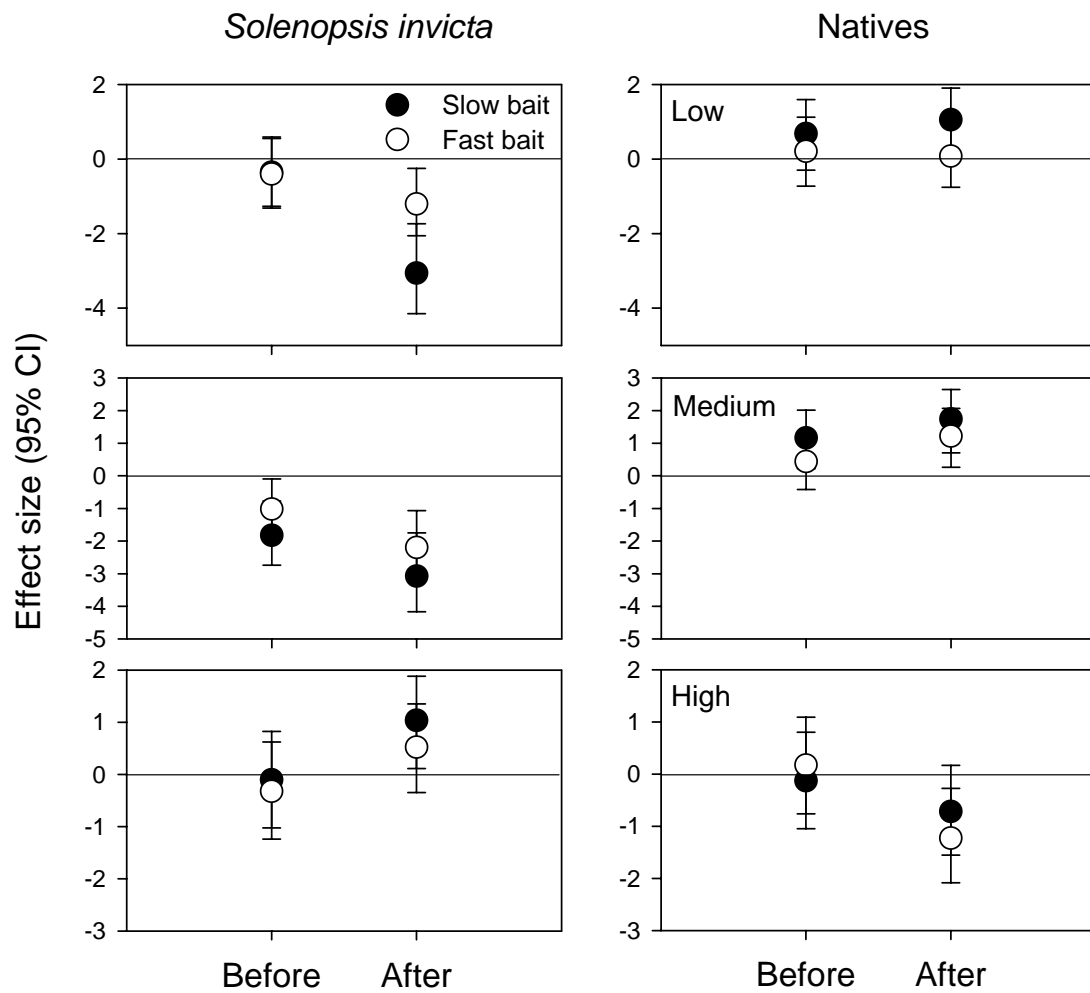


FIG. 34. Impact of baits on *S. invicta* and Natives (three densities, low, medium and high) based on food lures, FY 2007.

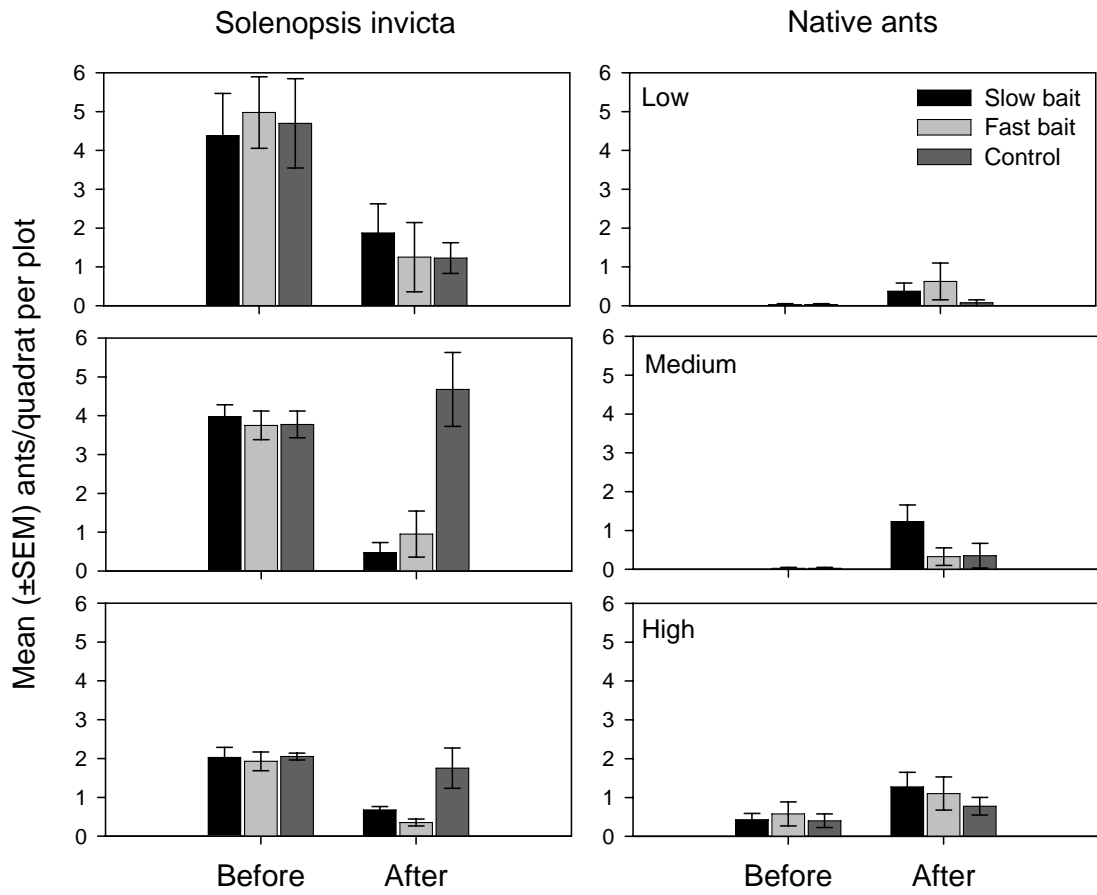


FIG. 35. Foraging behavior of *S. invicta* and Natives (three densities, low, medium and high) on placebo baits, FY 2006.

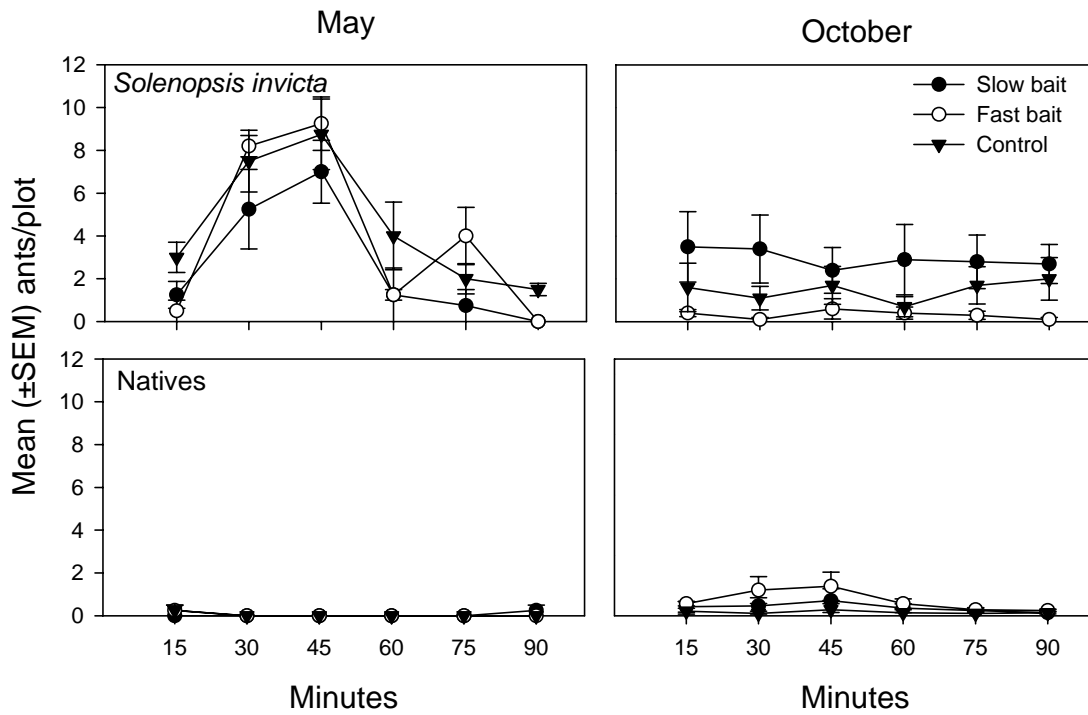


FIG. 36. Foraging behavior of *S. invicta* and Natives (Pruitt - Low native ant density) on placebo bait, before poison bait was applied and ~22 weeks after poison bait application. Observations taken at 15 min intervals following deployment of the placebo for a 90 minute period for the before and the after measurements, FY 2006.

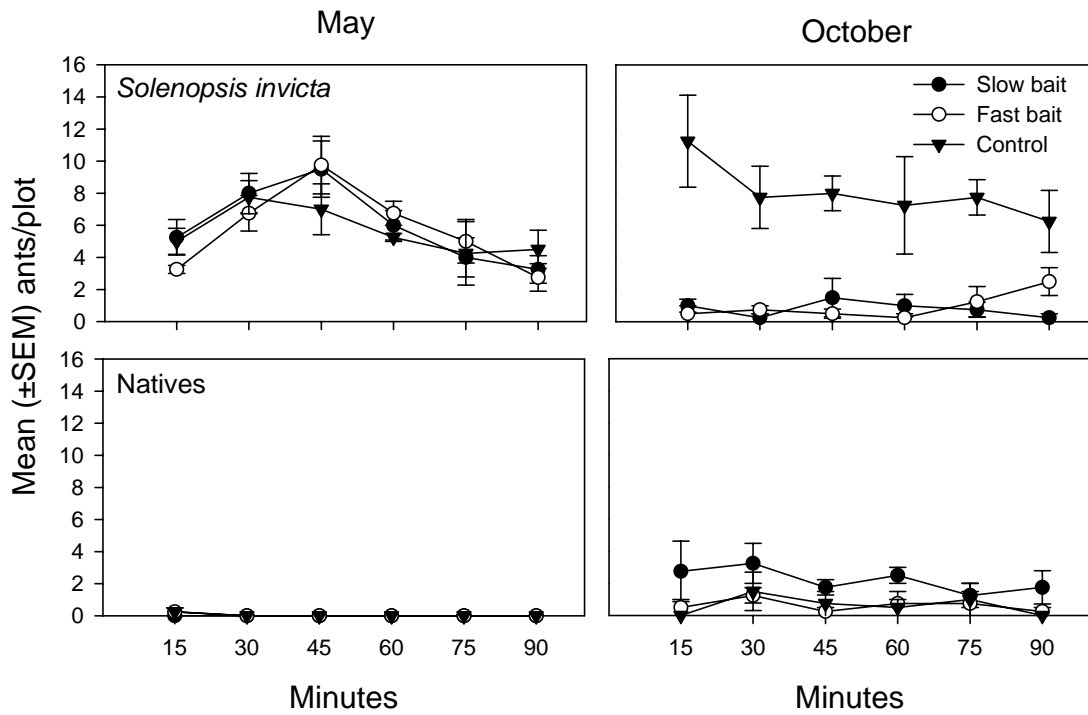


FIG. 37. Foraging behavior of *S. invicta* and Natives (Barr - Medium native ant density) on placebo bait, before poison bait was applied and ~22 weeks after poison bait application. Observations taken at 15 min intervals following deployment of the placebo for a 90 minute period for the before and the after measurements, FY 2006.

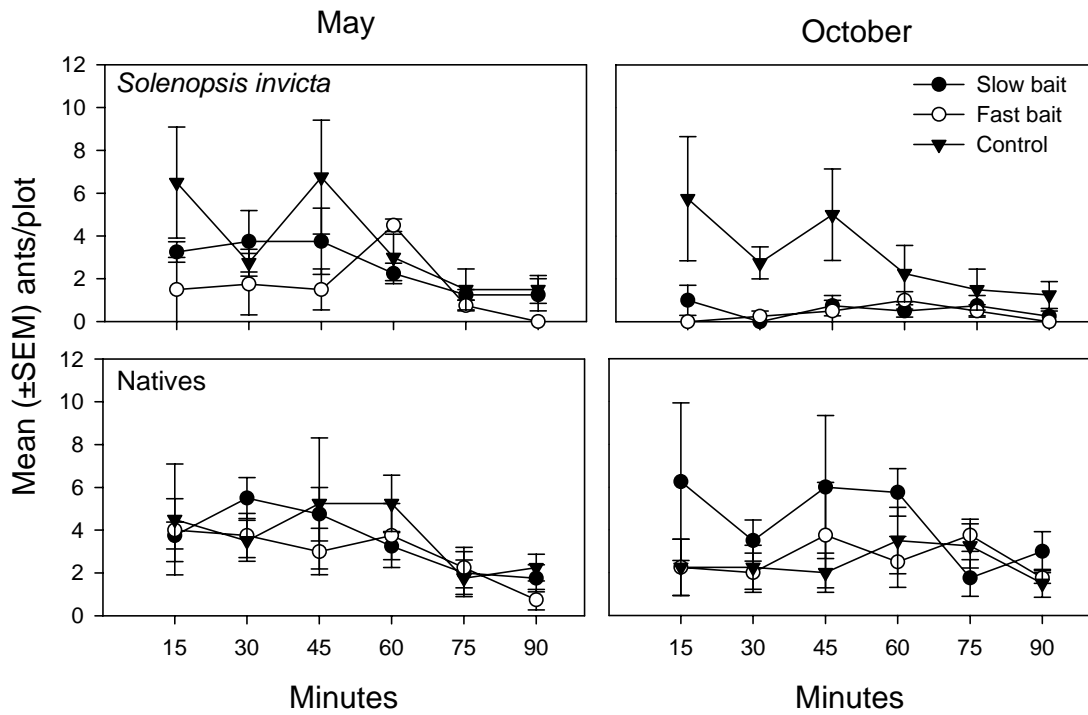


FIG. 38. Foraging behavior of *S. invicta* and Natives (C3 - High native ant density) on placebo bait, before poison bait was applied and ~22 weeks after poison bait application. Observations taken at 15 min intervals following deployment of the placebo for a 90 minute period for the before and the after measurements, FY 2006.

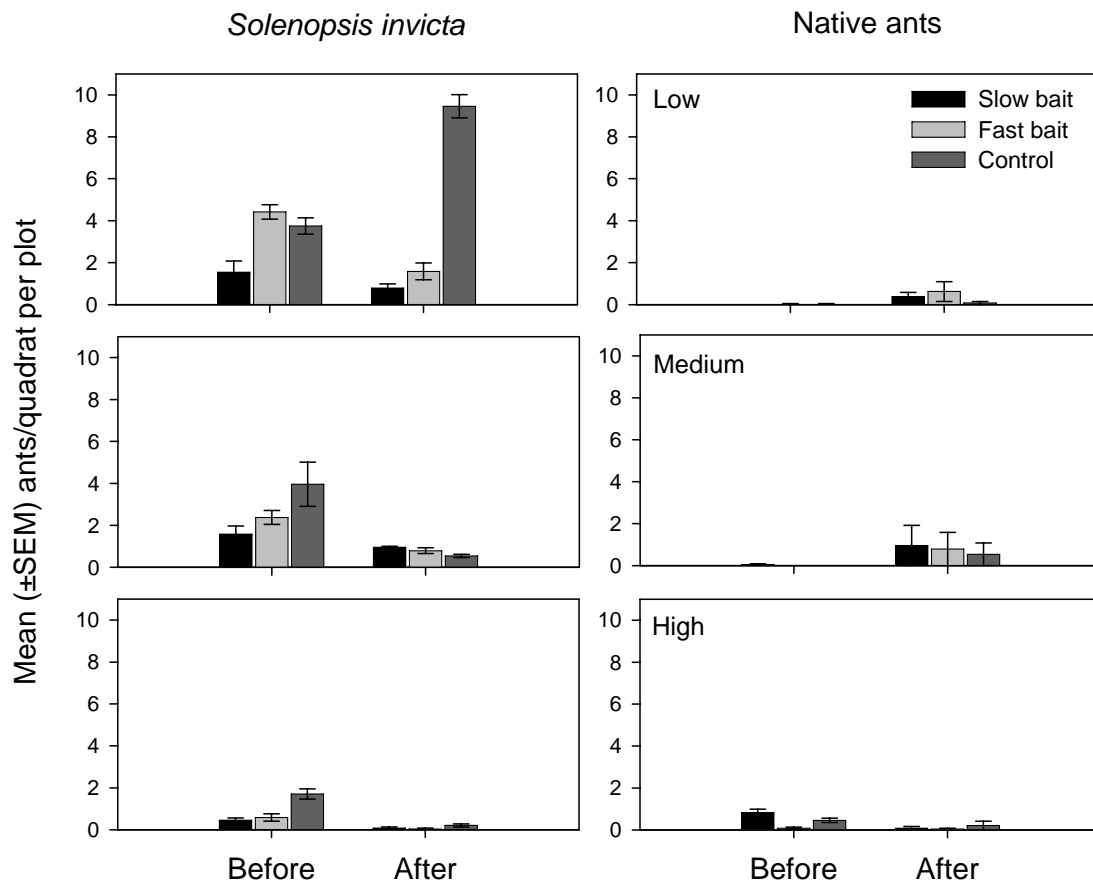


FIG. 39. Foraging behavior of *S. invicta* and Natives (three densities, low, medium and high) on placebo baits, FY 2007.



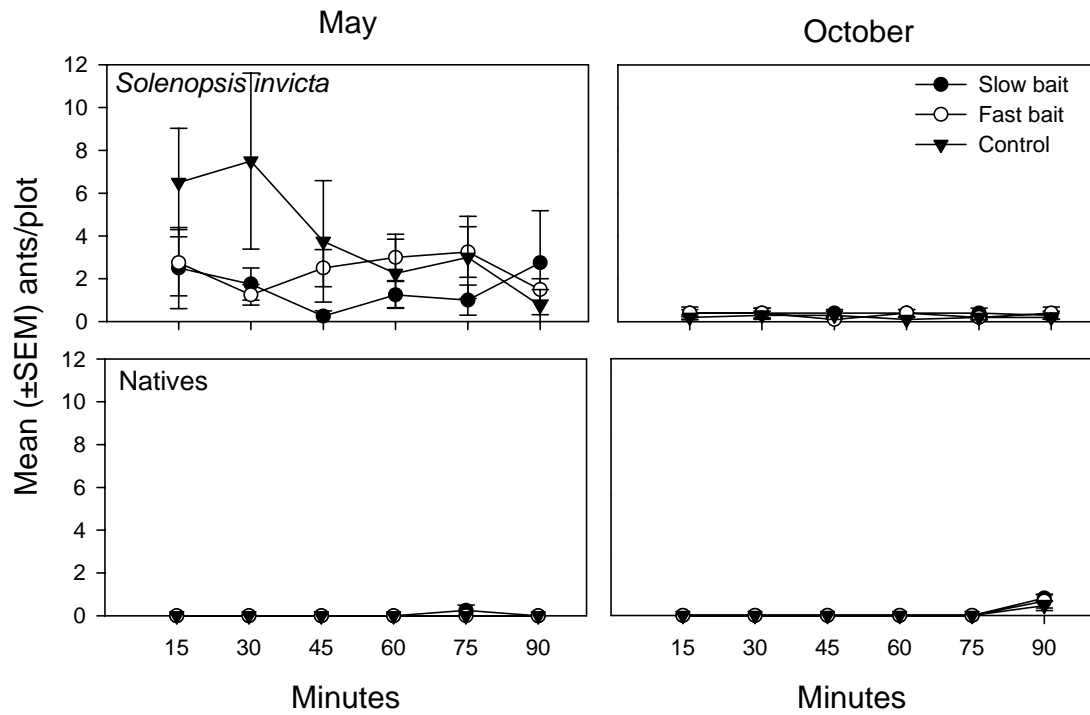


FIG. 40. Foraging behavior of *S. invicta* and Natives (Pruitt - Low native ant density) on placebo bait, before poison bait was applied and ~22 weeks after poison bait application. Observations taken at 15 min intervals following deployment of the placebo for a 90 minute period for the before and the after measurements, FY 2007.

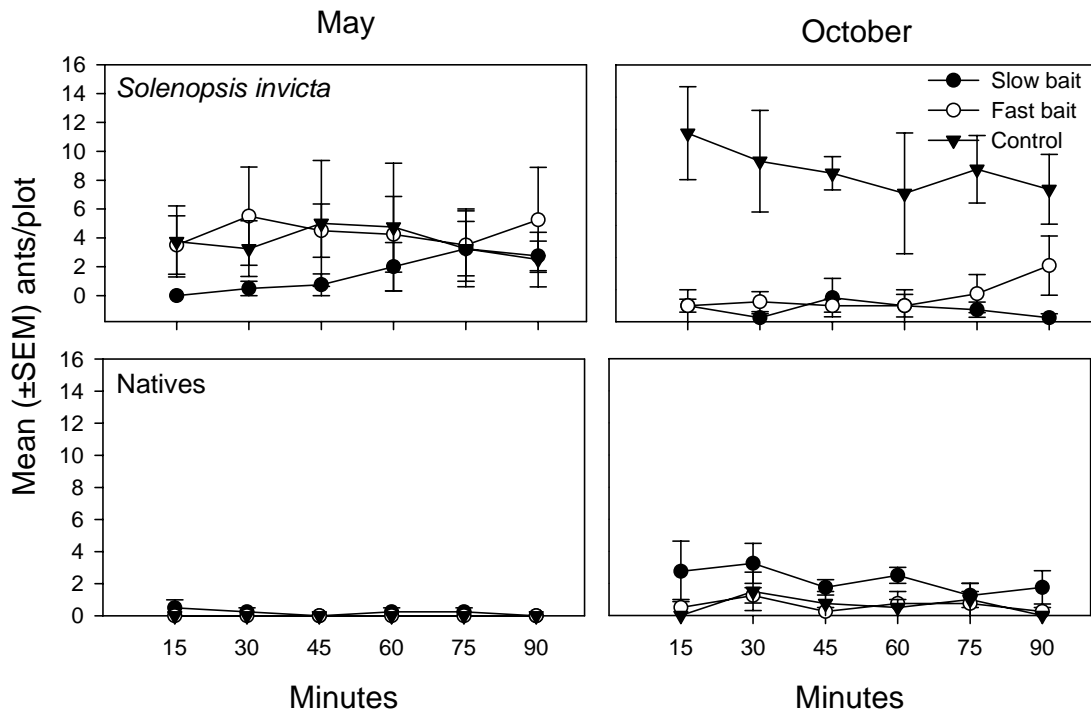


FIG. 41. Foraging behavior of *S. invicta* and Natives (Barr - Medium native ant density) on placebo bait, before poison bait was applied and ~22 weeks after poison bait application. Observations taken at 15 min intervals following deployment of the placebo for a 90 minute period for the before and the after measurements, FY 2007.

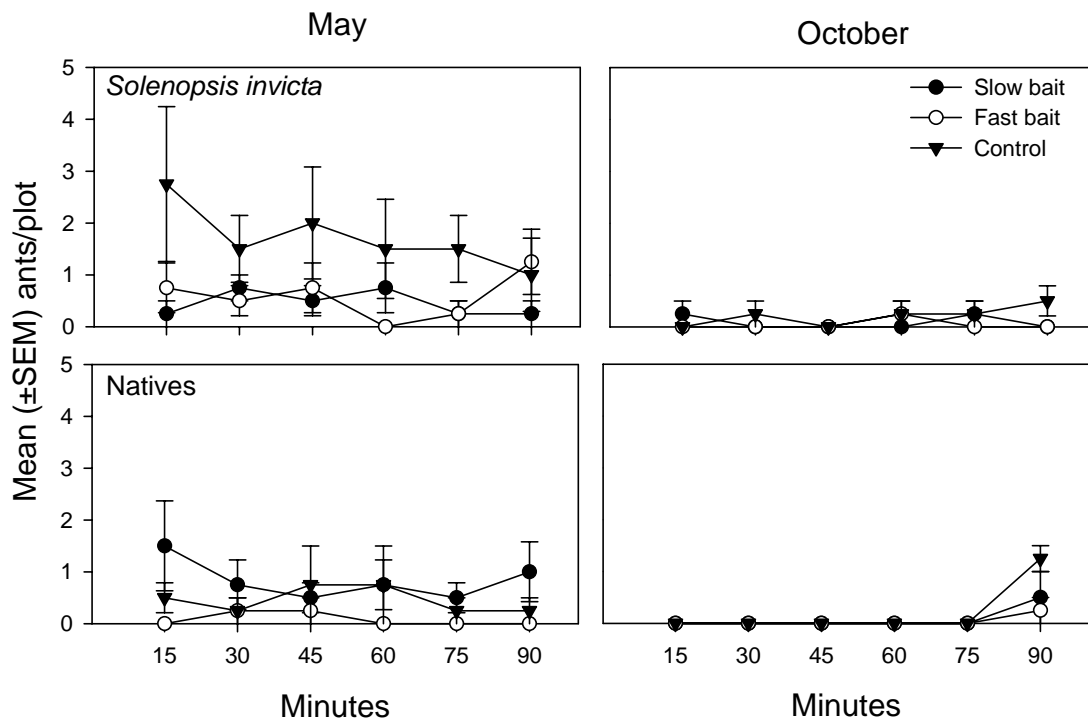


FIG. 42. Foraging behavior of *S. invicta* and Natives (C3 - High native ant density) on placebo bait, before poison bait was applied and ~22 weeks after poison bait application. Observations taken at 15 min intervals following deployment of the placebo for a 90 minute period for the before and the after measurements, FY 2007.

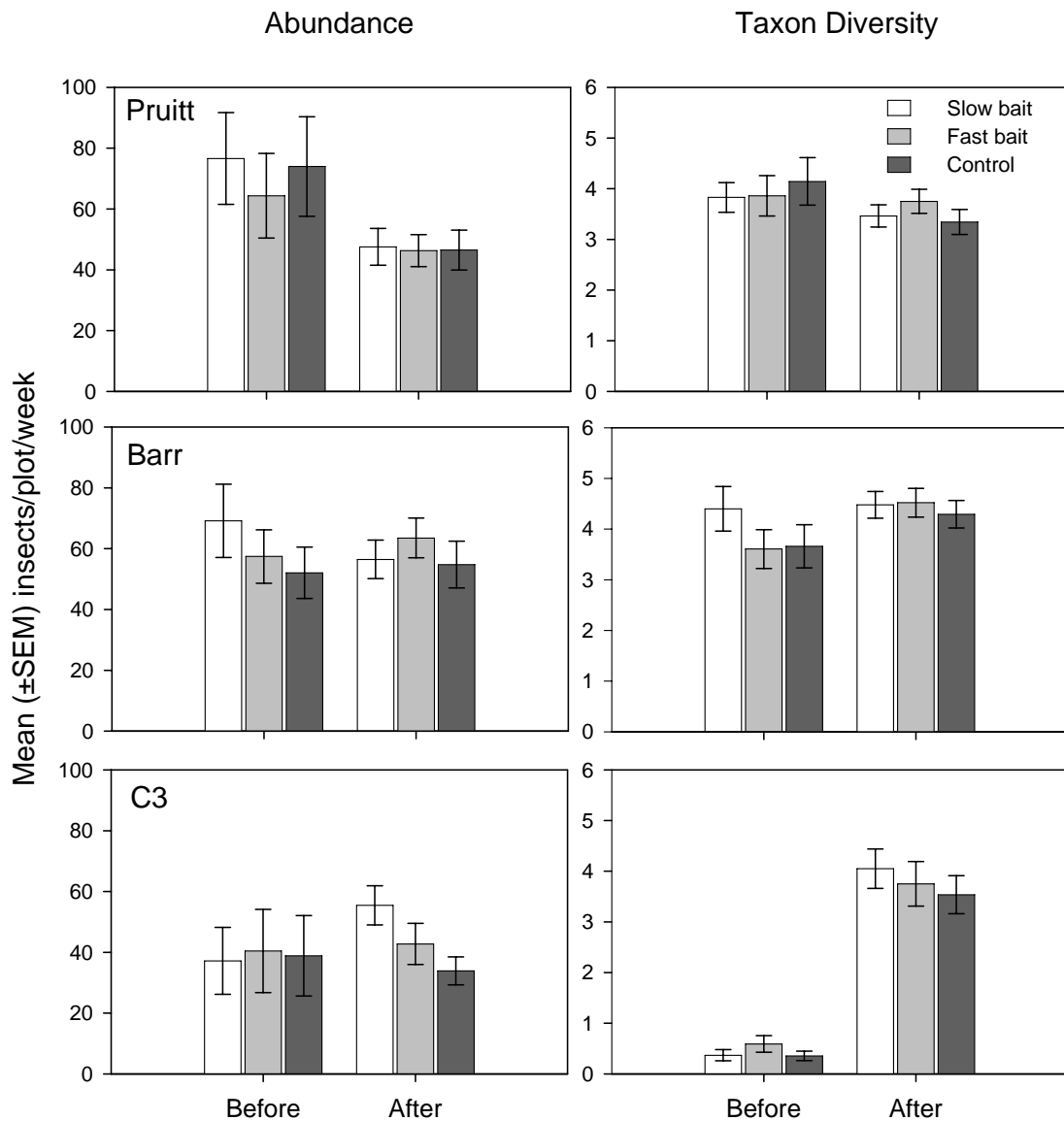


FIG. 43. Impact of insecticide baits and *S. invicta* reduction on relative insect abundance (non-ant) and taxon diversity (order level) based on pitfall traps across the three study sites, FY 2006.

## APPENDIX 2

## TABLES

TABLE 1. Sampling dates for each site before and after the treatment application. Treatments were applied at a rate of 0.68kg/ha using a Herd seeder (Model GT-77 ATV - Herd Seeder Co. Inc.) mounted on a utility vehicle (John Deere Gator TH 4x4, Deere & Company). Treatments were applied on May 16, 2006 and June 7, 2007 on “Barr” and May 17, 2006 and June 6, 2007 on “C3” and “Pruitt”.

2006				2007			
	Barr	C3	Pruitt		Barr	C3	Pruitt
Before	15-Mar	31-Mar	31-Mar	Before	9-Mar	2-Apr	2-Apr
	22-Mar	5-Apr	5-Apr		6-Apr	9-Apr	9-Apr
	30-Mar	12-Apr	12-Apr		13-Apr	25-Apr	25-Apr
	6-Apr	19-Apr	19-Apr		20-Apr	2-May	2-May
	13-Apr	26-Apr	26-Apr		27-Apr	9-May	9-May
	18-Apr	3-May	3-May		4-May	16-May	16-May
	28-Apr	11-May	11-May		11-May	23-May	24-May
	4-May				18-May	30-May	30-May
	9-May				25-May	6-Jun	6-Jun
					1-Jun		
After	23-May	24-May	24-May		7-Jun		
	30-May	31-May	31-May	After	22-Jun	21-Jun	21-Jun
	8-Jun	7-Jun	7-Jun		28-Jun	11-Jul	11-Jul
	15-Jun	14-Jun	16-Jun		6-Jul	17-Jul	17-Jul
	22-Jun	23-Jun	21-Jun		12-Jul	25-Jul	25-Jul
	29-Jun	28-Jun	28-Jun		18-Jul	1-Aug	1-Aug
	6-Jul	6-Jul	5-Jul		2-Aug	8-Aug	8-Aug
	13-Jul	12-Jul	12-Jul		9-Aug	15-Aug	15-Aug
	19-Jul	20-Jul	20-Jul		17-Aug	24-Aug	24-Aug
	27-Jul	26-Jul	26-Jul		25-Aug	30-Aug	30-Aug
	2-Aug	2-Aug	2-Aug		5-Oct	4-Oct	5-Oct
	30-Aug	9-Aug	10-Aug		30-Oct	1-Nov	1-Nov
	29-Sep	31-Aug	31-Aug				
	26-Oct	28-Sep	28-Sep				
		27-Oct					

TABLE 2. Ant species and abundance at “Pruitt” site by year among the different treatments, using pitfall traps.

Pitfall traps Species	2006						2007					
	Before			After			Before			After		
	Slow	Fast	Control	Slow	Fast	Control	Slow	Fast	Control	Slow	Fast	Control
<b>Dolichoderinae</b>												
<i>Dorymyrmex flavus</i> McCook	0	0	13	0	1	7	0	2	2	0	19	29
<i>Forelius pruinus</i> Roger	960	2367	2340	13259	20869	15783	761	768	332	4267	1647	1750
<b>Ecitoninae</b>												
<i>Ladibius coecus</i> Latreille	1	0	0	0	0	0	0	0	0	1	3	11
<i>Neivamyrmex opacithorax</i> Emery	0	0	0	0	6	0	0	0	0	0	6	0
<b>Formicinae</b>												
<i>Brachymyrmex depilis</i> Emery	0	0	2	56	10	34	0	0	2	3	1	1
<i>Paratrechina terricola</i> Buckley	122	66	93	414	391	656	122	60	49	337	388	210
<b>Myrmicinae</b>												
<i>Carebara longii</i> Wheeler	0	0	0	0	0	0	0	0	0	0	1	0
<i>Crematogaster laeviuscula</i> Mayr	0	0	1	0	0	0	0	3	1	10	3	7
<i>Cyphomyrmex wheeleri</i> Forel	0	0	1	243	14	97	1	0	0	0	2	0
<i>Monomorium minimum</i> Buckley	0	0	1	15	16	10	2	0	1	1	2	10
<i>Pheidole sp.</i>	3	13	60	9	6	170	3	1	37	92	17	16
<i>Pogonomyrmex barbatus</i> F. Smith	0	1	0	2	30	3	0	16	4	2	4	3
<i>Solenopsis invicta</i> Buren	0	0	1	1	6	108	0	1	9	1	7	72
<i>Solenopsis molesta</i> Say	4181	2984	6298	20109	2607	27077	550	461	1024	15103	23288	44535
<i>Strumigenys silvestrii</i> Emery	3	6	0	19	42	24	2	0	0	0	0	1
<i>Trachymyrmex sp.</i>	0	0	0	1	0	0	0	0	0	0	0	0
<b>Ponerinae</b>												
<i>Hypoponera opacior</i> Forel	1	1	1	0	2	0	0	0	0	1	2	0
Total number of specimens	5270	5437	8811	34128	24000	43969	1441	1312	1461	19818	25390	46645
Raw species richness	7	7	11	11	13	11	7	8	10	11	15	12

TABLE 3. Ant species and abundance at “Pruitt” site by year among the different treatments, using food lures.

Species	2006						2007					
	Before			After			Before			After		
	Slow	Fast	Control	Slow	Fast	Control	Slow	Fast	Control	Slow	Fast	Control
<b>Dolichoderinae</b>												
<i>Dorymyrmex flavus</i> McCook	2	0	0	151	70	0	0	0	0	0	0	0
<i>Forelius pruinosus</i> Roger	511	2540	971	11582	9372	2640	589	135	105	3484	696	745
<b>Formicinae</b>												
<i>Paratrechina terricola</i> Buckley	10	62	0	50	24	150	21	81	2	199	102	0
<b>Myrmicinae</b>												
<i>Crematogaster laeiuscula</i> Mayr	0	0	0	0	10	0	0	0	25	0	5	0
<i>Monomorium minimum</i> Buckley	20	252	210	0	350	50	0	2	2	2	51	0
<i>Pheidole sp.</i>	0	0	0	0	0	0	0	0	0	0	50	5
<i>Pogonomyrmex barbatus</i> F. Smith	0	0	0	0	1	16	0	0	25	0	0	0
<i>Solenopsis invicta</i> Buren	20520	18034	22344	8306	4018	17459	7772	7464	9261	8958	16201	24219
<i>Solenopsis molesta</i> Say	0	0	0	51	10	0	0	0	0	0	0	0
Total number of specimens	21063	20888	23525	20140	13855	20315	8382	7682	9420	12643	17105	24969
Raw species richness	5	4	3	5	8	5	3	4	6	4	6	3

TABLE 4. Ant species and abundance at “Barr” site by year among the different treatments, using pitfall traps.

Species	2006						2007					
	Before			After			Before			After		
	Slow	Fast	Control	Slow	Fast	Control	Slow	Fast	Control	Slow	Fast	Control
<b>Dolichoderinae</b>												
<i>Dorymyrmex flavus</i> McCook	150	23	8	198	90	225	104	106	199	237	45	217
<i>Forelius pruinosus</i> Roger	385	465	340	2127	850	1671	765	701	396	1851	739	4872
<i>Tapinoma sessile</i> Say	26	3	3	195	32	75	100	45	72	131	94	110
<b>Ecitoninae</b>												
<i>Neivamyrmex isodentatus</i> MacKay	0	0	0	1	0	0	0	0	0	0	1	0
<b>Formicinae</b>												
<i>Brachymyrmex depilis</i> Emery	0	0	0	12	1	10	0	0	1	0	0	0
<i>Camponotus pennsylvanicus</i> Degeer	0	0	0	1	1	0	0	1	0	0	0	1
<i>Camponotus sansabeanus</i> Buckley	0	0	0	0	0	1	1	0	1	0	0	0
<i>Camponotus sayi</i> Emery	0	0	0	1	0	0	0	0	0	0	1	0
<i>Paratrechina terricola</i> Buckley	48	63	84	316	510	564	147	309	315	271	691	313
<b>Myrmicinae</b>												
<i>Crematogaster laeviuscula</i> Mayr	1	4	0	0	16	10	0	18	1	5	7	1
<i>Cyphomyrmex rimosus</i> Spinola	0	0	2	27	46	106	0	1	0	2	8	7
<i>Cyphomyrmex wheeleri</i> Forel	15	7	4	54	29	41	10	7	7	11	4	12
<i>Monomorium minimum</i> Buckley	40	4	1	129	31	19	82	66	110	176	216	152
<i>Myrmecina americana</i> Emery	0	0	0	0	0	2	0	0	0	0	0	2
<i>Pheidole lamia</i> Wheeler	0	0	0	0	0	0	0	0	0	0	3	2
<i>Pheidole sp.</i>	0	12	1	1	101	30	7	85	10	3	202	25
<i>Solenopsis invicta</i> Buren	1730	861	974	4007	1313	6860						
<i>Solenopsis molesta</i> Say	2	1	2	6	19	32	0	16	0	10	37	31
<i>Strumigenys silvestrii</i> Emery	0	0	0	14	12	2	0	1	0	1	4	5
<b>Ponerinae</b>												
<i>Hypoponera opacior</i> Forel	0	0	0	1	4	3	1	0	0	0	13	
<i>Leptogenys elongata</i> Buckley	0	0	0	0	0	1	0	0	1	0	1	0
<b>Psuedomyrmecinae</b>												
<i>Pseudomyrmex ejectus</i> Smith	0	0	0	1	0	0	0	0	0	1	1	0



Total number of specimens	2397	1443	1419	7091	3055	9652	1217	1356	1113	2699	2067	5750
Raw species richness	9	10	10	17	15	17	9	12	11	12	17	14

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TABLE 5. Ant species and abundance at “Barr” site by year among the different treatments, using food lures.

Food lures Species	2006						2007					
	Before			After			Before			After		
	Slow	Fast	Control	Slow	Fast	Control	Slow	Fast	Control	Slow	Fast	Control
Dolichoderinae												
<i>Dorymyrmex flavus</i> McCook	0	200	0	374	177	110	50	0	0	96	0	0
<i>Forelius pruinosus</i> Roger	2775	1131	1217	6601	5511	1525	2261	1009	1310	3950	927	1045
Formicinae												
<i>Paratrechina terricola</i> Buckley	200	335	420	835	3234	51	120	172	0	873	1433	68
Myrmicinae												
<i>Crematogaster laeviuscula</i> Mayr	0	0	0	0	125	0	0	2	0	0	35	0
<i>Crematogaster sp. 2</i>	0	0	0	0	0	0	0	25	0	0	0	0
<i>Monomorium minimum</i> Buckley	100	527	150	2130	2655	210	819	393	200	1391	1461	0
<i>Pheidole sp.</i>	0	0	0	100	103	0	0	21	10	0	92	10
<i>Solenopsis geminata</i> Fabricius	0	0	0	0	2	0	0	0	0	0	0	0
<i>Solenopsis invicta</i> Buren	20446	21098	21195	13917	7528	35125	12964	15384	20320	9253	10400	25071
<i>Solenopsis molesta</i> Say	0	100	0	0	100	0	10	50	50	180	77	328
Total number of specimens	23521	23391	22982	23957	19435	37021	16224	17056	21890	15743	14425	26522
Raw species richness	4	6	4	6	9	5	6	8	5	6	7	5

TABLE 6. Ant species and abundance at “C3” site by year among the different treatments, using pitfall traps.

Species	2006						2007					
	Before			After			Before			After		
	Slow	Fast	Control	Slow	Fast	Control	Slow	Fast	Control	Slow	Fast	Control
<b>Dolichoderinae</b>												
<i>Dorymyrmex flavus</i> McCook	11	27	77	41	14	186	8	1	18	160	14	97
<i>Forelius pruinosus</i> Roger	6291	1248	1134	73393	18417	17401	11786	1314	363	15474	4758	2070
<i>Tapinoma sessile</i> Say	0	0	0	0	0	0	0	0	0	1	0	0
<b>Ecitoninae</b>												
<i>Labidus coecus</i> Latreille	0	0	0	0	0	1	3	1	0	0	0	0
<i>Neivamyrmex texanus</i> Watkins	21	0	2	6	2	7	0	0	0	0	0	0
<b>Formicinae</b>												
<i>Brachymyrmex depilis</i> Emery	0	0	0	11	19	4	1	2	1	4	4	0
<i>Camponotus sansabeanus</i> Buckley	2	0	0	0	0	0	1	0	0	3	0	0
<i>Camponotus sayi</i> Emery	4	1	0	2	2	0	10	0	0	3	1	1
<i>Paratrechina terricola</i> Buckley	94	197	91	514	463	425	95	97	42	921	700	388
<b>Myrmicinae</b>												
<i>Carebara longii</i> Wheeler	0	0	0	0	0	0	0	0	0	0	3	0
<i>Crematogaster laeviuscula</i> Mayr	28	5	1	58	65	0	39	10	0	82	62	2
<i>Crematogaster sp. 2</i>	2	20	0	9	52	4	8	20	1	4	20	0
<i>Cyphomyrmex wheeleri</i> Forel	2	1	1	7	15	7	2	2	0	5	2	4
<i>Leptothorax obturator</i> Wheeler	0	0	0	0	0	0	0	0	0	1	0	0
<i>Monomorium minimum</i> Buckley	130	80	90	276	45	272	96	21	68	625	427	577
<i>Myrmecina americana</i> Emery	0	0	2	1	1	3	0	0	1	0	1	0
<i>Pheidole lamia</i> Wheeler	0	0	0	0	0	0	0	0	0	0	0	17
<i>Pheidole sp. 1</i>	2	19	2	11	18	30	1	5	7	5	4	18
<i>Pheidole hyatti</i> Emery	0	0	0	0	6	7	0	0	4	1	1	0
<i>Pheidole sp. 3</i>	0	0	0	0	0	2	0	0	0	0	0	0
<i>Pogonomyrmex barbatus</i> F. Smith	0	0	1	0	0	0	0	0	0	0	0	0
<i>Pyramica margaritae</i> Forel	0	0	0	0	0	2	0	0	0	0	0	0
<i>Solenopsis invicta</i> Buren	3329	1757	4188	9668	5120	32007	324	446	3211	6142	7688	40683
<i>Solenopsis molesta</i> Say	2	25	25	80	113	62	6	116	18	90	307	67

<i>Strumigenys silvestrii</i> Emery	2	2	0	31	7	23	0	0	0	4	3	4
<i>Trachymyrmex sp.</i>	0	0	0	2	0	2	1	0	0	1	0	0
Ponerinae												
<i>Hypoponera opacior</i> Forel	0	0	0	0	0	1	0	0	0	2	0	0
Total number of specimens	9920	3382	5614	84110	24359	50446	12381	2035	3734	23527	13995	43928
Raw species richness	14	12	12	16	16	19	15	12	11	18	16	12

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TABLE 7. Ant species and abundance at “C3” site by year among the different treatments, using food lures.

Species	2006						2007					
	Before			After			Before			After		
	Slow	Fast	Control	Slow	Fast	Control	Slow	Fast	Control	Slow	Fast	Control
<b>Dolichoderinae</b>												
<i>Dorymyrmex flavus</i> McCook	0	50	0	192	425	124	0	0	0	20	0	0
<i>Forelius pruinosus</i> Roger	4120	5239	6592	10023	8502	11825	978	2318	1960	920	102	1516
<i>Forelius sp. 2</i>	0	0	101	0	50	50	0	0	0	0	0	0
<b>Formicinae</b>												
<i>Brachymyrmex depilis</i> Emery	0	0	0	0	0	0	0	0	1	0	0	0
<i>Paratrechina terricola</i> Buckley	1	1	0	157	401	71	93	30	136	554	279	804
<b>Myrmicinae</b>												
<i>Crematogaster laeiuscula</i> Mayr	208	0	0	550	50	110	102	65	10	0	8	85
<i>Monomorium minimum</i> Buckley	50	360	1730	1460	842	781	355	302	917	30	1	395
<i>Pheidole sp.</i>	0	15	0	0	0	10	0	0	0	10	0	0
<i>Solenopsis invicta</i> Buren	17072	15431	13833	17015	11164	12396	10834	10018	11247	18302	16575	12889
<i>Solenopsis molesta</i> Say	0	0	0	0	5	0	10	0	76	21	0	200
Total number of specimens	21451	21096	22256	29397	21439	25367	12372	12733	14347	19857	16965	15889
Raw species richness	5	6	4	6	8	8	6	5	7	7	5	6

TABLE 8. Linear Mixed Models results for effects of treatments on the relative diversity of ants based on pitfall traps, excluding *Solenopsis invicta*, during 2006.

		Richness					
		Before			After		
<b>Pruitt</b>	Source	<i>df</i> *	F	<i>P</i>	<i>df</i>	F	<i>P</i>
	Treatments	2,8	0.581	0.580	2,109	2.810	0.065
	Week	5,2	1.538	0.385	12,15	1.028	0.471
	Treatment x Week	0,2	3.319	0.178	24,15	0.560	0.901
<b>Barr</b>	Source	<i>df</i>	F	<i>P</i>	<i>df</i>	F	<i>P</i>
	Treatments	2,9	0.259	0.777	2,9	0.259	0.777
	Week	7,12	9.432	0.000	7,12	9.432	0.000
	Treatment x Week	14,12	1.118	0.427	14,12	1.118	0.427
<b>C3</b>	Source	<i>df</i>	F	<i>P</i>	<i>df</i>	F	<i>P</i>
	Treatments	2,11	0.053	0.949	2,94	1.659	0.196
	Week	5,16	9.784	0.000	12,17	4.218	0.003
	Treatment x Week	10,16	0.639	0.762	24,17	0.455	0.963

\* Denotes degrees of freedom of Linear Mixed Model (numerator, denominator).

TABLE 9. Linear Mixed Models results for effects of treatments on the relative diversity of ants based on pitfall traps, excluding *Solenopsis invicta* during 2007.

		Richness					
		Before			After		
<b>Pruitt</b>	Source	<i>df</i> *	F	<i>P</i>	<i>df</i>	F	<i>P</i>
	Treatments	2,8	0.288	0.756	2,79	1.433	0.245
	Week	8,12	3.787	0.018	9,16	5.515	0.001
	Treatment x Week	16,12	1.567	0.213	18,16	0.495	0.925
<b>Barr</b>	Source	<i>df</i>	F	<i>P</i>	<i>df</i>	F	<i>P</i>
	Treatments	2,8	0.120	0.888	2,93	1.066	0.348
	Week	9,13	4.096	0.010	10,15	8.047	0.000
	Treatment x Week	18,13	2.789	0.030	20,15	0.692	0.783
<b>C3</b>	Source	<i>df</i>	F	<i>P</i>	<i>df</i>	F	<i>P</i>
	Treatments	2,9	0.197	0.825	2,76	2.599	0.081
	Week	8,15	5.555	0.002	9,18	26.778	0.000
	Treatment x Week	16,15	0.497	0.912	18,18	0.560	0.887

\* Denotes degrees of freedom of Linear Mixed Model (numerator, denominator).

TABLE 10. Co-occurrence patterns of ants on the three study sites before and after the bait treatment. Mean columns under observed indicates the estimated C-scores of the model. Communities with high co-occurrence should frequently fail to reject the null hypothesis. Small variances would mean that treatments are strikingly similar to one another in the level of co-occurrence observed.

	2006					2007				
	Observed		Estimated		<i>P</i> -value	Observed		Estimated		<i>P</i> -value
	Mean	Variance	Mean	Variance		Mean	Variance	Mean	Variance	
<b>Pruitt</b>										
Before	0.82	0.09	0.69	0.07	0.25	0.32	0.12	0.44	0.07	0.14
After	0.38	0.07	0.33	0.03	0.05	0.49	0.02	0.51	0.03	0.44
<b>Barr</b>										
Before	0.41	0.05	0.41	0.03	0.24	0.41	0.05	0.21	0.03	0.24
After	0.25	0.01	0.31	0.02	0.83	0.44	0.00	0.44	0.03	0.91
<b>C3</b>										
Before	0.44	0.00	0.46	0.01	0.60	0.43	0.05	0.48	0.05	0.20
After	0.45	0.00	0.46	0.01	0.76	0.37	0.01	0.50	0.03	0.68



TABLE 11. Linear Mixed Models results for effects of treatments on the relative abundance and behavioral dominance of *Solenopsis invicta* and native ants during 2006 in “Pruitt”.

Pruitt		<i>Solenopsis invicta</i>						Natives					
		Before			After			Before			After		
	Source	df*	F	P	df	F	P	df	F	P	df	F	P
Food Lures	Treatments	2,13	0.366	0.700	2,14	43.965	0.000	2,3	1.213	0.411	2,4	0.581	0.597
	Week	6,12	23.193	0.000	13,14	17.561	0.000	6,	1.455	**	13,13	3.530	0.013
	Treatment x Week	12,12	1.266	0.342	26,14	4.966	0.001	12,	0.866	**	26,13	1.235	0.349
Pitfall traps	Treatments	2,38	4.153	0.023	2,25	1.282	0.295	2,25	0.612	0.550	2,14	0.068	0.935
	Week	5,21	3.437	0.020	12,6	2.347	0.133	5,14	1.099	0.404	12,16	1.333	0.286
	Treatment x Week	10,21	0.365	0.949	24,6	1.672	0.250	10,14	0.344	0.952	24,16	1.143	0.394
Quadrats	Treatments	2,9	2.271	0.156	2,56	38.934	0.000	2,9	1.321	0.312	2,4	3.789	0.101
	Time	5,11	0.244	0.935	5,7	19.037	0.000	5,11	0.516	0.760	5,5	9.643	0.011
	Treatment x Time	10,11	1.320	0.325	10,7	5.708	0.014	10,11	0.868	0.583	10,5	3.836	0.067

\* Denotes degrees of freedom of Linear Mixed Model (numerator, denominator).

\*\* Not enough information to perform the Linear Mixed Model, test inconclusive.

TABLE 12. Linear Mixed Models results for effects of treatments on the relative abundance and behavioral dominance of *Solenopsis invicta* and native ants during 2006 in “Barr”.

<b>Barr</b>		<i>Solenopsis invicta</i>						Natives					
		Before			After			Before			After		
	Source	<i>df</i>	F	<i>P</i>	<i>df</i>	F	<i>P</i>	<i>df</i>	F	<i>P</i>	<i>df</i>	F	<i>P</i>
Food Lures	Treatments	2,9	0.092	0.913	2,10	68.978	0.000	2,	0.473	**	2,8	2.389	0.147
	Week	8,13	16.723	0.000	14,13	14.662	0.000	8,30	4.037	0.002	14,12	9.272	0.000
	Treatment x Week	16,13	1.220	0.359	28,13	11.351	0.000	16,30	0.912	0.565	28,12	3.141	0.019
Pitfall traps	Treatments	2,10	2.644	0.118	2,36	12.270	0.000	2,39	0.532	0.591	2,20	1.205	0.320
	Week	7,15	5.180	0.003	12,17	13.139	0.000	7,12	5.315	0.005	12,14	8.324	0.000
	Treatment x Week	14,15	1.228	0.346	24,17	7.196	0.000	14,12	0.527	0.875	24,14	1.206	0.362
Quadrats	Treatments	2,51	0.542	0.585	2,8	14.607	0.002	2,9	1.000	0.405	2,	1.472	**
	Time	5,28	11.757	0.000	5,13	0.216	0.950	1,9	1.000	0.343	5,19	1.437	0.255
	Treatment x Time	10,28	1.711	0.128	10,13	0.694	0.716	2,9	1.000	0.405	10,19	1.307	0.294

\* Denotes degrees of freedom of Linear Mixed Model (numerator, denominator).

\*\* Not enough information to perform the Linear Mixed Model, test inconclusive.

TABLE 13. Linear Mixed Models results for effects of treatments on the relative abundance and behavioral dominance of *Solenopsis invicta* and native ants during 2006 in “C3”.

C3		<i>Solenopsis invicta</i>						Natives					
		Before			After			Before			After		
	Source	df*	F	P	df	F	P	df	F	P	df	F	P
Food Lures	Treatments	2,16	2.302	0.132	2,43	1.325	0.276	2,52	3.186	0.049	2,77	0.703	0.498
	Week	6,13	38.639	0.000	15,13	17.039	0.000	6,29	26.958	0.000	15,13	12.609	0.000
	Treatment x Week	12,13	1.479	0.241	30,16	4.993	0.001	12,29	1.019	0.458	30,13	0.760	0.744
Pitfall traps	Treatments	2,30	4.120	0.026	2,8	5.496	0.029	2,14	2.185	0.148	2,7	1.743	0.243
	Week	5,23	8.587	0.000	12,15	3.439	0.012	5,13	1.294	0.323	12,3	3.102	0.158
	Treatment x Week	10,23	1.908	0.096	24,15	1.875	0.102	10,13	0.813	0.622	24,3	1.268	0.467
Quadrats	Treatments	2,215	2.506	0.084	2,44	14.516	0.000	2,16	1.021	0.382	2,11	8.334	0.006
	Time	5,20	9.890	0.000	5,17	1.380	0.279	5,12	8.545	0.001	5,16	1.203	0.351
	Treatment x Time	10,20	0.942	0.518	10,17	1.963	0.104	10,12	1.003	0.489	10,16	1.825	0.136

\* Denotes degrees of freedom of Linear Mixed Model (numerator, denominator).

TABLE 14. Linear Mixed Models results for effects of treatments on the relative abundance and behavioral dominance of *Solenopsis invicta* and native ants during 2007 in “Pruitt”.

Pruitt		<i>Solenopsis invicta</i>						Natives					
		Before			After			Before			After		
	Source	<i>df</i> *	F	<i>P</i>	<i>df</i>	F	<i>P</i>	<i>df</i>	F	<i>P</i>	<i>df</i>	F	<i>P</i>
Food Lures	Treatments	2,17	1.189	0.327	2,85	36.331	0.000	2,35	2.922	0.067	2,32	4.577	0.018
	Week	8,17	17.360	0.000	10,18	8.848	0.000	8,13	1.964	0.134	10,15	2.203	0.077
	Treatment x Week	16,17	1.691	0.142	20,18	0.846	0.643	16,13	1.474	0.242	20,15	1.263	0.322
Pitfall traps	Treatments	2,36	6.267	0.005	2,52	6.826	0.002	2,44	0.769	0.469	2,44	3.191	0.051
	Week	8,16	12.360	0.000	9,15	7.945	0.000	8,30	2.780	0.019	9,22	2.519	0.037
	Treatment x Week	16,16	1.308	0.298	18,15	2.068	0.080	16,30	0.377	0.979	18,22	0.788	0.693
Quadrats	Treatments	2,10	1.137	0.357	2,8	0.346	0.717	2,13	0.928	0.420	2,13	0.480	0.629
	Time	5,13	0.870	0.526	5,8	0.188	0.959	5,45	0.206	0.958	5,45	4.235	0.003
	Treatment x Time	10,13	1.285	0.327	10,8	0.748	0.673	10,45	0.206	0.995	10,45	0.106	1.000

\* Denotes degrees of freedom of Linear Mixed Model (numerator, denominator).

TABLE 15. Linear Mixed Models results for effects of treatments on the relative abundance and behavioral dominance of *Solenopsis invicta* and native ants during 2007 in “Barr”.

<b>Barr</b>		<i>Solenopsis invicta</i>						Natives					
		<b>Before</b>			<b>After</b>			<b>Before</b>			<b>After</b>		
	Source	<i>df</i> *	F	<i>P</i>	<i>df</i>	F	<i>P</i>	<i>df</i>	F	<i>P</i>	<i>df</i>	F	<i>P</i>
Food Lures	Treatments	2,9	3.743	0.066	2,77	151.423	0.000	2,528	2.057	0.129	2,60	12.555	0.000
	Week	10,13	5.963	0.002	11,17	38.862	0.000	10,19	5.746	0.000	11,21	4.423	0.002
	Treatment x Week	20,13	1.997	0.098	22,17	7.743	0.000	20,19	0.833	0.656	22,21	1.629	0.133
Pitfall traps	Treatments	2,24	8.062	0.002	2,9	0.855	0.455	2,14	0.114	0.893	2,23	2.359	0.117
	Week	9,18	4.106	0.005	10,16	3.751	0.009	9,15	3.444	0.015	10,16	4.541	0.004
	Treatment x Week	18,18	1.272	0.305	20,16	2.197	0.057	18,15	1.464	0.225	20,16	1.095	0.431
Quadrats	Treatments	2,10	0.585	0.574	2,8	14.607	0.002	0,	**	**	2,	1.472	**
	Time	5,12	1.121	0.399	5,13	0.216	0.950	3,36	1.160	0.338	5,19	1.437	0.255
	Treatment x Time	10,12	0.724	0.691	10,13	0.694	0.716	6,27	1.200	0.336	10,19	1.307	0.294

\* Denotes degrees of freedom of Linear Mixed Model (numerator, denominator).

TABLE 16. Linear Mixed Models results for effects of treatments on the relative abundance and behavioral dominance of *Solenopsis invicta* and native ants during 2007 in “C3”.

C3		<i>Solenopsis invicta</i>						Natives					
		Before			After			Before			After		
	Source	df*	F	P	df	F	P	df	F	P	df	F	P
Food Lures	Treatments	2,18	0.292	0.750	2,85	2.740	0.070	2,42	0.446	0.643	2,44	11.167	0.000
	Week	8,12	12.138	0.000	10,17	3.353	0.013	8,16	5.727	0.001	10,14	4.687	0.004
	Treatment x Week	16,13	0.751	0.711	20,17	5.806	0.000	16,16	0.948	0.541	20,14	1.815	0.123
Pitfall traps	Treatments	2,6	2.354	0.165	2,23	37.718	0.000	2,12	0.489	0.625	2,19	5.473	0.013
	Week	8,18	2.711	0.037	9,18	9.696	0.000	8,11	1.484	0.264	9,11	4.482	0.011
	Treatment x Week	16,18	1.414	0.238	18,18	3.907	0.003	16,11	1.200	0.385	18,11	0.964	0.543
Quadrats	Treatments	2,10	1.472	0.257	2,8	0.346	0.717	2,13	0.928	0.420	2,13	0.480	0.629
	Time	5,19	1.437	0.255	5,8	0.188	0.959	5,45	0.206	0.958	5,45	4.235	0.003
	Treatment x Time	10,19	1.307	0.294	10,8	0.748	0.673	10,45	0.206	0.995	10,45	0.106	1.000

\* Denotes degrees of freedom of Linear Mixed Model (numerator, denominator).

TABLE 17. List of ant species observed at quadrats discovering and collecting placebo baits at “Pruitt”.

Species	2006						2007					
	Before			After			Before			After		
	Slow	Fast	Control	Slow	Fast	Control	Slow	Fast	Control	Slow	Fast	Control
Dolichoderinae												
<i>Forelius sp.</i>	41	14	5	28	20	1	1	0	0	1	0	0
Myrmicinae												
<i>Monomorium minimum</i>	1	1	6	0	0	0	0	0	0	0	0	0
<i>Solenopsis invicta Buren</i>	19	177	88	38	8	119	38	57	95	23	19	13
Total number of specimens	61	192	99	66	28	120	39	57	95	24	19	13
Raw species richness	3	3	3	2	2	2	2	1	1	2	1	1

TABLE 18. List of ant species observed at quadrats discovering and collecting placebo baits at “Barr”.

Species	2006						2007					
	Before			After			Before			After		
	Slow	Fast	Control	Slow	Fast	Control	Slow	Fast	Control	Slow	Fast	Control
Dolichoderinae												
<i>Dorymyrmex flavus</i>	0	0	1	2	1	0	0	0	0	5	0	0
<i>Forelius sp.</i>	0	0	0	46	5	14	0	0	0	0	0	0
Formicinae												
<i>Paratrechina terricola</i>	0	0	0	4	2	1	0	0	0	0	0	0
Myrmicinae												
<i>Monomorium minimum</i>	0	0	0	1	7	0	0	0	0	0	0	0
<i>Solenopsis invicta</i> Buren	125	122	151	19	38	227	29	100	90	16	9	13
Total number of specimens	125	122	152	72	53	242	29	100	90	21	9	13
Raw species richness	1	1	2	5	5	3	1	1	1	2	1	1



TABLE 19. List of ant species observed at quadrats discovering and collecting placebo baits at “C3”.

Species	2006						2007					
	Before			After			Before			After		
	Slow	Fast	Control	Slow	Fast	Control	Slow	Fast	Control	Slow	Fast	Control
Dolichoderinae												
<i>Dorymyrmex flavus</i>	0	0	0	0	0	0	0	0	2	0	0	0
<i>Forelius sp.</i>	19	42	18	51	50	31	20	2	9	0	0	0
Myrmicinae												
<i>Monomorium minimum</i>	3	0	0	0	0	0	0	0	0	0	0	0
<i>Solenopsis invicta</i> Buren	62	28	88	13	9	70	11	14	41	2	1	5
Total number of specimens	84	70	106	64	59	101	31	16	50	2	1	5
Raw species richness	3	2	2	2	2	2	2	2	2	1	1	1

TABLE 20. Linear Mixed Models results for effects of treatments on the relative abundance and taxa diversity of insect during 2006.

		Abundance						Taxa					
		Before			After			Before			After		
<b>Pruitt</b>	Source	<i>df</i> *	F	<i>P</i>	<i>df</i>	F	<i>P</i>	<i>df</i>	F	<i>P</i>	<i>df</i>	F	<i>P</i>
	Treatments	2,17	0.974	0.397	2,11	0.046	0.956	2,10	0.570	0.582	2,12	1.380	0.287
	Week	5,16	53.511	0.000	11,17	30.311	0.000	5,13	19.846	0.000	11,16	23.715	0.000
	Treatment x Week	10,16	0.393	0.931	22,17	0.405	0.977	10,13	0.521	0.848	22,17	1.003	0.505
<b>Barr</b>	Source	<i>df</i>	F	<i>P</i>	<i>df</i>	F	<i>P</i>	<i>df</i>	F	<i>P</i>	<i>df</i>	F	<i>P</i>
	Treatments	2,17	1.753	0.202	2,98	2.861	0.062	2,8	2.814	0.114	2,77	0.963	0.386
	Week	7,15	42.891	0.000	12,16	17.149	0.000	7,15	16.607	0.000	12,17	10.992	0.000
	Treatment x Week	14,15	1.860	0.175	24,16	1.669	0.144	14,15	1.055	0.456	24,17	1.029	0.485
<b>C3</b>	Source	<i>df</i>	F	<i>P</i>	<i>df</i>	F	<i>P</i>	<i>df</i>	F	<i>P</i>	<i>df</i>	F	<i>P</i>
	Treatments	2,19	0.074	0.929	2,82	12.102	0.000	2,33	0.593	0.558	2,3515	0.443	0.642
	Week	5,12	47.074	0.000	13,10	137.829	0.000	5,17	148.639	0.000	13,34	8103.160	0.000
	Treatment x Week	10,12	0.360	0.944	25,9	1.849	0.163	10,17	0.769	0.657	25,14	1.277	0.317

\* Denotes degrees of freedom of Linear Mixed Model (numerator, denominator).

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## Selected Publications

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