

## ABSTRACT

BEDNAR, DAVID MICHAEL. *Pachycondyla* (= *Brachyponera*) *chinensis* Predation on *Reticulitermes virginicus* and Competition with *Aphaenogaster rudis*. (Under the direction of Dr. Jules Silverman.)

Invasive ant species have general diet and nest requirements, which facilitate their establishment in novel habitats and their dominance over many native ants. The Asian needle ant, *Pachycondyla* (= *Brachyponera*) *chinensis*, native throughout Australasia was introduced to the Southeastern United States where it has become established in woodland habitats, nests in close proximity to and consumes subterranean termites (Rhinotermitidae). We illustrate that *P. chinensis* do not occur in habitats lacking Rhinotermitidae and suggest that subterranean termites are critical for *P. chinensis* success in new habitats. We demonstrate that *P. chinensis* is a general termite feeder, retrieving *Reticulitermes virginicus* five times more often than other potential prey near *P. chinensis* colonies. Odors produced by *R. virginicus* workers, as well as other potential prey, attract *P. chinensis*. Furthermore, *P. chinensis* occupy *R. virginicus* nests in the lab and field and display behaviors that facilitate capture of *R. virginicus* workers and soldiers. Termites are an abundant, high quality, renewable food supply, in many ways similar to the hemipteran honeydew exploited by most other invasive ant species. We conclude that the behavior of *P. chinensis* in the presence of termites increases their competitive abilities in natural areas where they have been introduced.

*Aphaenogaster rudis* (complex) worker abundance is strongly negatively correlated with *P. chinensis* worker abundance, suggesting that the latter species competitively displaces the former. We examined the competitive ability of *P. chinensis* to better

understand the mechanisms that may, in part, be responsible for the displacement of *A. rudis* (complex) nests by *P. chinensis* colonies. *Pachycondyla chinensis* dominated *A. rudis* (complex) workers in direct worker-worker and whole colony interactions, whereby most *A. rudis* (complex) are killed by *P. chinensis* workers. Furthermore, while both *A. rudis* (complex) and *P. chinensis* nests co-occur with and prey upon subterranean termites (*Reticulitermes spp.*), *P. chinensis* foragers discover nests, kill, and retrieve workers and soldiers of *R. virginicus* more quickly than *A. rudis* (complex) foragers. We demonstrated that termite defense systems such as soldier ratio and wood barriers have little if any hindering effects on *P. chinensis* forager ability to prey on termites. *Pachycondyla chinensis* appear to be the better termite predator. Consequently, *P. chinensis* competitive ability is enhanced in the presence of termites. When *P. chinensis* workers discover *A. rudis* workers foraging they kill them. When *P. chinensis* workers encounter an *A. rudis* nest, they kill them. Termites affect these interactions between *P. chinensis* and *A. rudis* workers by increasing the number of ant forager interactions.

*Pachycondyla* (= *Brachyponera*) *chinensis* Predation on *Reticulitermes virginicus* and  
Competition with *Aphaenogaster rudis*

by  
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## **DEDICATION**

I would like to dedicate this to my parents who, always told me to let the bugs go before I went to bed, and to my wife who, encouraged me to follow my passion. Also, to my nephews, James Thomas and Watson Houck, possible future entomologists.

## BIOGRAPHY

David Michael Bednar was born November 12<sup>th</sup>, 1984 in Klamath Falls, Oregon to Kevin and April Bednar. His parents encouraged him in every endeavor, while teaching him to enjoy nature at every opportunity. David attended a small high school in Condon, Oregon and graduated from a class of 21 students. He spent his summers working various jobs at game bird farms, wheat farms, and the Gilliam County road department. It was at the road department that David was first introduced to the idea of biological control of weeds using insects. He took advantage of every opportunity the school could offer him, allowing him to graduate as a five-sport varsity athlete.

After graduating from Condon High School, David attended Pacific University in Forest Grove, Oregon. The small private college allowed David to compete on the varsity wrestling, track, and cross country teams while at the same time staying active in club sports like crew and ultimate (frisbee). The small class sizes at Pacific created a close relationship between students and professors. Three professors in particular inspired David to pursue entomology. However, it was at Pacific where he met Maria, who, later became his wife and encouraged him to follow his passion.

Ed Alkaslassy, worked with David as his majors advisor, but it was his previous work as a museum curator of the insect collection that intrigued him most. Dr. Brook Swanson was a young new professor who introduced David to insect morphology and physiology. Dr. Stacey Halpern probably influenced David the most offering him an internship in Florida the summer after his Junior year. David studied the oviposition behavior of false Colorado potato beetles (*Leptinotarsa juncta*), and immediately became passionate about studying insects.

David wrote his senior capstone on the invasion biology of argentine ants (*Linepithema humile*). In his research for the project David became interested in working with Dr. Jules Silverman.

David was accepted to North Carolina State University, under the advisement of Dr. Jules Silverman immediately after receiving his Bachelor of Science in May of 2007. After finishing his Master's of Science David will continue his work in Entomology. Under the advisement of Dr. Fred Hain, David will work toward a Phd studying the balm wooly adelgid.

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When one works in a lab as excellent as I was privileged, thanks do not seem enough. I will humbly try to include all those who made this work possible, and grant them my deepest, sincere, appreciation.

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## CHAPTER 1

### **Use of termites (*Reticulitermes virginicus*) as a springboard in the invasive success of a predatory ant (*Pachycondyla* (= *Brachyponera*) *chinensis*)**

In general, those ant species that have become successfully established outside of their native range share rather indiscriminate food preferences and vagile nesting habits (McGlynn 1999, Holway et al. 2002). Ants occupy a variety of ecological niches throughout the world's biospheres. Ant nesting structures range from simple subterranean soil tunnels (Tschinkel 2003), to symbiotic plants (Janzen 1973), and even include mobile bivouacs (Anderson et al. 2002). Ant feeding habits are equally diverse, ranging from obligate specialist predators (Wheeler 1936) to fungus growing farmers (Quinlan and Cherrett 1979), or general scavengers (Hölldobler and Wilson 1990, Lodge 1993, McGlynn 1999). Diets of the most destructive invasive ants, such as *Linepithema humile*, *Solenopsis invicta*, *Pheidole megacephala*, *Wasmannia auropunctata*, and *Anoplolepis longipes* include arthropod prey, hemipteran honeydew, and extrafloral nectar (Way 1963, Bach 1991, Abril et al. 2007, Tillberg et al. 2007, Helms and Vinson 2008). It seems reasonable to assume it is unlikely that ant species having narrow food and/or nesting preferences, e.g. arboreal ants have low establishment rates in new habitats presumably from a lack of suitable habitat. Flexibility, for some ants, could allow them to adapt to novel diets within their region of introduction (Suarez et al. 2005).

Many successful invasive ants rely on hemipteran honeydew as a springboard to their invasion success. Hemipteran honeydew is nutritive, abundant, and available throughout most of the year (year-round in some climates). This food source allows ants, such as *L.*

*humile*, to grow massive colonies, and expand their territories (Morales 2000, Helms and Vinson 2002, Lach 2005, Grover et al. 2007, Tillberg et al. 2007, Helms and Vinson 2008, Rowles and Silverman 2009). Termites present an excellent candidate as a springboard for invasive predatory ants. Termites can be as abundant as 10,000 individuals/m<sup>2</sup> (Eggleton et al. 1996), they are available year-round, extremely nutritive (Sogbesan and Ugwumba 2008), and create suitable nest space for many ant species (Dejean et al. 1996).

*Pachycondyla* (= *Brachyponera*) *chinensis*, native throughout Australasia (Smith 1934, Wheeler 1936, Bolton 1995), and one of the most common native woodland ants throughout Japan (Kubota et al. 2003), is now regarded as an invasive pest in parts of the eastern U.S. (Guénard unpublished data). Recent evidence indicates that several native ant species in southeastern U.S. deciduous forests are negatively impacted by *P. chinensis* and it has been suggested that one mechanism by which *P. chinensis* influences other species is through competition for nest sites and food (Guénard unpublished data). Nests of *P. chinensis* occur on the forest floor within decayed logs. Subterranean termites in the genus *Reticulitermes* also frequently occupy these logs. Throughout their introduced range *P. chinensis* workers have been observed nesting with and carrying *Reticulitermes* spp. workers in their mandibles (Zungoli et al. 2005, Nelder et al. 2006). In Japan, where *P. chinensis* occur naturally, they have even been referred to as a “termite-hunter” (Matsuura 2002). Most evidence concerning the diet of *P. chinensis*, is anecdotal, however, being restricted to a few observations, where they are described as generalists or scavengers (Smith 1934).

Several other *Pachycondyla* spp. including: *P. commutata*, *P. laevigata*, and *P. marginata*, prey exclusively upon different termite species (Wheeler 1936, Mill 1984,

Agbogba 1992, Leal and Oliveira 1995, Garcia Perez et al. 1997, Corbara and Dejean 2000, Bayliss and Fielding 2002). Here, we test the hypothesis that the invasive ant, *P. chinensis*, is a generalized termite predator, which prefers termites as food but will accept other prey items. We test this hypothesis by examining the behavior of *P. chinensis* workers towards *Reticulitermes virginicus* (Isoptera: Rhinotermitidae) workers and other arthropods in decayed logs occupied by *P. chinensis* nests. We also examine the movement of *P. chinensis* workers into nests occupied by colonies of *R. virginicus*.

## **Methods and Materials**

### **Distribution of *P. chinensis* and termite prey**

To determine whether termites provide introduced *P. chinensis* with a food source and/or nesting substrate, we tested if the occurrence of *P. chinensis* was limited by the distribution of termites. Distribution maps have been created for ants based on presence/absence data (Guénard et al. 2009). These maps are helpful in predicting the spread of potential threats like *P. chinensis*. If termites are a requirement in the success of these invaders a demonstration of overlapping geographic range is necessary.

We used presence and absence data for political regions throughout the world for *Pachycondyla chinensis* (Davies et al. 2003, Dunn et al. 2005). We performed a literature search using ISI Web of Science<sup>sm</sup> and produced presence absence data for termite species belonging to the genera *Reticulitermes* and *Coptotermes*, which have both been cited as nesting with *P. chinensis* (Nelder et al. 2006). Rhinotermitidae include wood-feeding termites with the widest global distribution (Eggleton 2000). They also include serious pest species such as *Coptotermes* and *Reticulitermes* (Eggleton 2000). Maps were created using

arcmap software displaying the sympatric and allopatric distributions of the two termite genera *Reticulitermes* and *Coptotermes* with *Pachycondyla* (= *Brachyponera*) *chinensis*.

Termite data were collected from the following sources: Weesner 1965, Rebello and Martius 1994, Medeiros et al. 1999, Eggleton 2000, Lo and Evans 2007, Bourguignon et al. 2009, Constantino 2002, Marini and Mantovani 2002, Scheffrahn et al. 2004, Szalanski et al. 2004, Ye et al. 2004, Austin et al. 2006, Park et al. 2006, Luchetti et al. 2007, Achetasham and Akhtar 2008, Neoh and Lee 2009, Yeap et al. 2009.

### **Collection of colonies and species determination**

*Pachycondyla chinensis* were identified in the field using site identification characteristics, including pupal cocoons. *Pachycondyla chinensis* workers demonstrate a smooth, dark shiny appearance, with lighter coloring of the terminal abdominal segments, distal leg segments, and mandibles, and large compound eyes. In the lab we verified other characters such as the deep mesonotal suture encircling a protruding mesonotum, and antennal scapes that extend beyond the posterior of the head. They are separated from other invasive *Pachycondyla* by the absence of metatarsal bristles on the hind legs (Smith 1934).

*Reticulitermes virginicus* and *R. flavipes* cannot be distinguished by site identification alone. Although, typically we noted *R. virginicus* colonies contained more workers than *R. flavipes*. Termites that were captured in the field were identified using molecular techniques described by Szalanski et al. (2003). DNA was extracted from workers preserved in alcohol using Chelex-100, as this was a less intensive process than other methods (Walsh et al. 1991). A portion of the COII region of the mtDNA was then amplified using PCR. TaqI

polymerase was used to digest the DNA, and electrophoresis with a 2.5% agarose gel was used to compare field specimens to known *R. flavipes* DNA profiles (Szalanski et al. 2003).

We collected one *P. chinensis* and one *R. virginicus* nest from Lake Johnson State Park (N 35.757966 W -78.713762) and one nest from Schenck Memorial Forest (N 35.81737 and W -78.72778) in May 2008. Schenck Memorial forest is dominated by *Pinus taeda* (10 – 70 yr). Lake Johnson State Park is dominated by oak (*Quercus sp.*, ca. 85-yr) and pine (*Pinus sp.*).

We separated *P. chinensis* individuals (ca. 2,000) and *R. virginicus* individuals (ca. 50,000) from the two field-collected logs, one from each site. *Pachycondyla chinensis* stock colonies were kept from each site in open plastic containers (76x76x6.4cm) with plastic petri dish nests (140x15mm) containing moistened plaster and provided water and food (10 *R. virginicus* workers/day plus artificial diet (Bhatkar and Whitcomb 1970) *ad libitum*. *Reticulitermes virginicus* stock colonies were held in plastic containers (46x35x12cm) with a moist cellulose matrix plus fragments of the wood they were extracted from. We maintained all insects at 26±0.7°C and 80% RH with a 12:12 L/D cycle.

***Pachycondyla* (=Brachyponera) chinensis acceptance of potential prey, including *R. virginicus***

A number of different arthropod species, including *R. virginicus*, were collected from logs with *P. chinensis*. We determined whether any or all of these arthropods might comprise the diet of *P. chinensis*. Arthropods were removed from field debris and identified to order, and to family and genus, where possible. Potential prey that were recovered included Curculionidae adults, Pyralidae larvae, Mycetophilidae larvae, Diplopoda, Machilidae,

*Reticulitermes virginicus*, Elateridae larvae, Trogossitidae adults, Chilopoda, *Parcoblatta* spp. nymphs, and Collembola. Ants were excluded, under the assumption that ants were more likely to be predators/competitors than prey.

We placed individuals from the same source colony (n=5; 1 queen, 25 workers, and 5 brood) of *P. chinensis* in a plastic Fluon-coated tray (25cmx18cmx5cm). Each tray was provisioned with a nest that consisted of a petri dish (90x10mm) with moistened cellulose. Ants were fed artificial diet (Bhatkar and Whitcomb 1970) for 24 hrs, after which, diet was removed and experimental prey were introduced. Of the arthropods recovered from logs with *P. chinensis*, we only tested those present year-round during our collections; *Reticulitermes virginicus* (workers), Elateridae (larvae), Trogossitidae (adults), Chilopoda, *Parcoblatta* spp. (nymphs), and Collembola.

We performed a no-choice experiment with these putative arthropod prey species. We introduced a different prey type on each of five consecutive days. For example, on day one, we introduced one *Parcoblatta* nymph to each of the five *P. chinensis* colonies, Chilopoda on day 2, etc. Prey not retrieved within ten-minutes were removed. We introduced the next prey item after 40 min. We recorded several observations of interactions between the ants and prey (List of observations in Table 2.1). We determined that acceptance of prey only included those that were returned to the ant nest within ten minutes. Results of prey retrieved were compared to instances where they were not using a Chi<sup>2</sup> analysis in JMP (SAS-v7.0 2007).

**Table 1.1 Variables used to assess *P. chinensis* behavior towards potential prey.**

Variable	Description
$D_t$	Time prey is discovered in seconds
E	Total number of encounters with prey
A	Maximum aggression score (0=no discovery, 1=ignore, 2=avoid, 3=biting/lunging, 4=stinging)
K	0=prey not killed in 600s; 1=prey killed
$K_t$	Time point prey is envenomed - $D_t$
R	0=prey not retrieved in 600s; 1=prey retrieved
$R_t$	Time point prey item is retrieved - $K_t$
F	Number of workers foraging before prey is introduced
G	Number of ants participating in group retrieval

***Pachycondyla* (= *Brachyponera*) *chinensis* detection of *R. virginicus* and other prey olfactory cues**

We evaluated *P. chinensis* worker response to *R. virginicus* worker scent in a Y-tube olfactometer. Three Petri dishes (50x9mm) were connected via a plastic Y-tube (2.2cmx3mmID, 60°angle). We placed a termite worker in either the “right” or “left” Petri dish (n=30, 15 per side). We then introduced one *P. chinensis* worker to the “center” Petri dish, and covered the dishes with their lids. We recorded the direction of travel (right or left) by the ant from the center Petri dish. We considered that a choice was made when the end of the ant’s abdomen passed the intersection of the tube. Thus, we avoided recording “investigating” behavior, where ants placed their head in the intersection and antennated without proceeding. Ants, termites, and Y-tubes were replaced once a choice was recorded. We repeated this procedure using *Parcoblatta* nymphs (n=20, 10 per side), and collembolans (n=20, 10 per side) crossed with *R. virginicus* workers as well as cellulose powder fed on by termites (ca. 1g dry wt.) crossed with autoclaved cellulose (ca. 1g dry wt.) (n=20, 10 per side). We first determined there were no differences between right left orientation using A Chi<sup>2</sup> analysis comparing instances of ant worker movement to the right or left. We then pooled right and left handed results to compare the number of choices made by *P. chinensis* workers toward termite workers/cellulose powder (fed on by termites) versus nothing, *Parcoblatta* nymphs, collembolans, or cellulose powder (autoclaved) using a Chi<sup>2</sup> analysis using JMP (SAS-v7.0 2007).

## ***Pachycondyla* (=Brachyponera) chinensis nest choice assays**

### Termite monitoring stations:

We used termite-monitoring stations in laboratory and field experiments to determine if *P. chinensis* moved into substrate containing termites. Each TMS contained a wooden block (southern yellow pine (*Pinus taeda* L.) in the base, and a termite inspection cartridge (TIC) filled with cellulose in the space above the wooden block (Figure 2.1a). Subterranean termites (Rhinotermitidae) consume the cellulose matrix and the wooden block, as well as nest within the wooden block (Rojas and Morales-Ramos 2001).

### Lab assay:

Two stations were placed 10cm apart in a plastic container (46cmx35cmx17cm) containing sandy soil (9660cm<sup>3</sup>). We removed the cellulose matrix from each station, then exposed 10 of these cellulose matrices to 1000 *R. virginicus* each, for 30 days. The cellulose matrices with the termites were kept in separate containers until 24hrs prior to the experiment. *Pachycondyla* (=Brachyponera) *chinensis* (n=100, 1 queens, and 5 brood) were housed in Petri dishes (95mmx15mm) containing moist plaster and covered with red cellophane.

Each of 10 replicate containers had one cellulose matrix with termites and one w/o termites in the station, with a divider between the stations to prevent termite movement between cellulose matrices. After 24 hrs, we placed the plaster filled Petri dish with the *P. chinensis* colony between the two stations (Figure 1.1b). Preliminary experiments revealed that *P. chinensis* move out of a plaster nest into a cellulose matrix. Using instantaneous sampling, we recorded the position of worker ants within each container hourly on the first

day (10am - 4pm) and then once daily (10am) for five days. The measure obtained through this type of sampling is the proportion of sample points during which the behavior is occurring; for our experiment it would be expressed as:

$$\frac{P}{E},$$
 where P is the number of ants observed in a particular location and E is the

number of sampling events. The resultant number has no units, but is a score of frequency for that behavior (Martin and Bateson 1993). We analyzed these data using a Friedman test where frequency data were scored between locations and the Chi<sup>2</sup> test statistic was calculated by hand (Ambrose et al. 2002).

#### Field assay 1:

We identified seven logs containing *P. chinensis* nests at three locations: Research Triangle Park (RTP) (1 log), Walnut Creek (WC) (3 logs), and Schenck Memorial Forest (S) (3 logs). On March 1, 2009, we buried monitoring stations in the soil parallel to each log 30cm apart for a total of 10 stations per log, 5 stations on each side (Figure 1.1b). Logs at each site were spaced at least 100m apart, while a minimum of 6.7km separated the three sites. Stations were inspected weekly for ants and/or termites for three months. After which time, the cellulose portions of the stations were returned to the laboratory and thoroughly inspected.

#### Field assay 2:

After inspection of cellulose matrices (n=70) from field assay 1, we found no arthropods. We then moistened the cellulose in all stations with distilled water. One-half of these (n=35) were each exposed to 1000 *R. virginicus* workers for 30 days. Termite-excavated galleries and termite workers were evident within the cellulose matrix by day 30.

The *R. virginicus*-infested and un-infested cellulose matrices were returned to the field and inserted within the stations as in field assay 1 (figure 1.1b). In contrast to the previous assay, five of the stations on one side of the log contained termites while those on the other side did not. The stations with and without termites were assigned randomly. We recorded the presence or absence, as well as any visible counts of termites and ants in each station daily, for 3 days. The proportion of instances where ants were present in stations containing termites was compared to those without termites and analyzed using a one-way ANOVA in JMP (SAS-v7.0 2007).

### **Foraging strategies of *P. chinensis* workers in the presence and absence of termites**

We developed a laboratory assay allowing for direct observation of *P. chinensis* worker responses to *R. virginicus* workers and soldiers within their nest. Two hundred *R. virginicus* (198 workers plus 2 soldiers) were housed in Petri dish nests (60mm x 15mm) containing moist pine sawdust. These nests were connected to an empty Petri dish (50mm x 9mm) by plastic tubing (10mm x 2mmID). *Pachycondyla chinensis* (100 workers, 1 queen, and 10 brood) were housed in a moist plaster-filled Petri dish (100mm x 15mm), which was attached to the above empty Petri dish (60mm x 15mm) on one end and another empty Petri dish distal to the termite nest (Figure 1.2). This dish was provisioned with 25% sucrose solution and freshly killed cockroaches, *Blattella germanica*, *ad libitum*. Access of *P. chinensis* workers to the sawdust nest was prevented for 24 hours prior to the observation period by blocking the tube entering the dishes with and without termites.

We recorded the behavior of *P. chinensis* to sawdust-filled nests with and without *R. virginicus*. Observations of *P. chinensis* contact with or entering the termite (or empty) nest

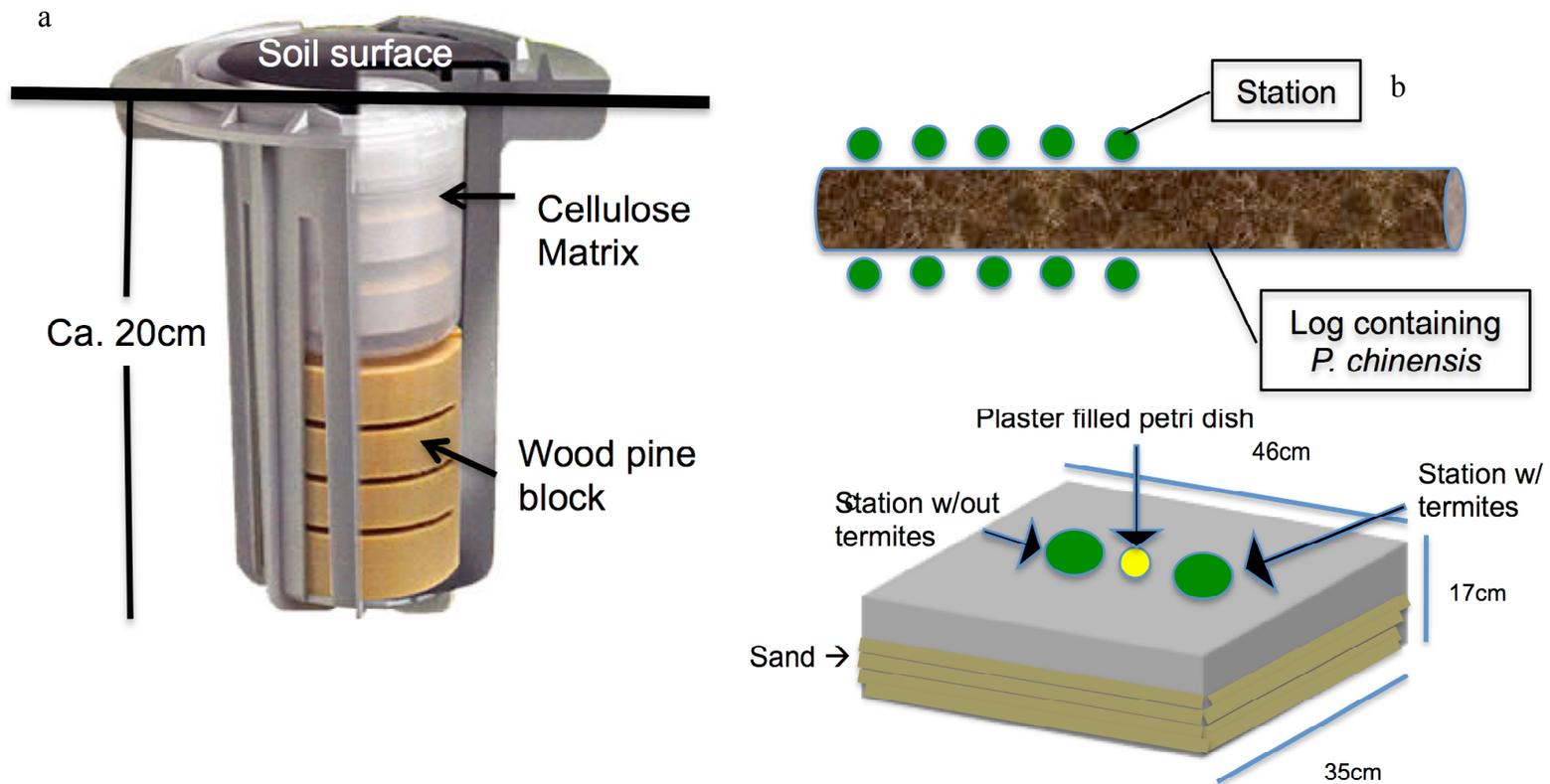


Figure 1.1 a) Station housing *R. virginicus* workers. b) Diagrammatic representation of how stations were oriented around a log containing a *P. chinensis* nest in field assay 1 and 2. c) Arrangement of plaster Petri dish and station housing termites in lab assay to monitor the movement of *P. chinensis* towards nesting substrate with and w/o *R. virginicus* in the laboratory.

entrance were recorded for four minutes every hour for 24 hours. We considered that *P. chinensis* 'entered' the nest when the entire length of her body was within the nest. If an ant contacted the nest entrance and immediately entered the nest we only recorded this as an entering event. We used repeated measures analysis of variance in JMP (SAS-v7.0 2007) to compare the number of contact and entering events between the treatments and controls over time.

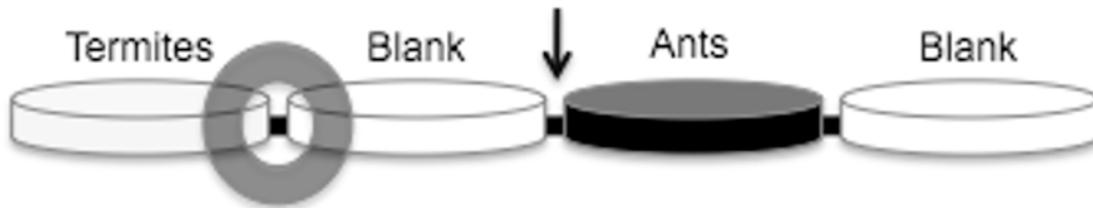


Figure 1.2 Diagram of lab set-up for observing ant foraging behavior in the presence and absence of termites. The arrow indicates the blockage point during the 24-hour acclimation period. The doughnut indicates the position where observations were recorded.

## Results

### Distribution of *P. chinensis* and termite prey

Distribution maps created for *Pachycondyla* (= *Brachyponera*) *chinensis* and two genera within Rhinotermitidae show *P. chinensis* only occur sympatrically with these termites. These termite genera also occur in areas not occupied by *P. chinensis*. (Figure 1.3).

### *Pachycondyla chinensis* acceptance of potential prey including *R. virginicus*

*Pachycondyla chinensis* workers retrieved all the *R. virginicus* presented. Significantly fewer ( $\chi^2=95.8$ ,  $p<0.0001$ ) Collembola (20%), *Parcoblatta spp.* (8%), Chilopoda (16%), Trogossitidae (0%), and Elateridae (0%) were retrieved.

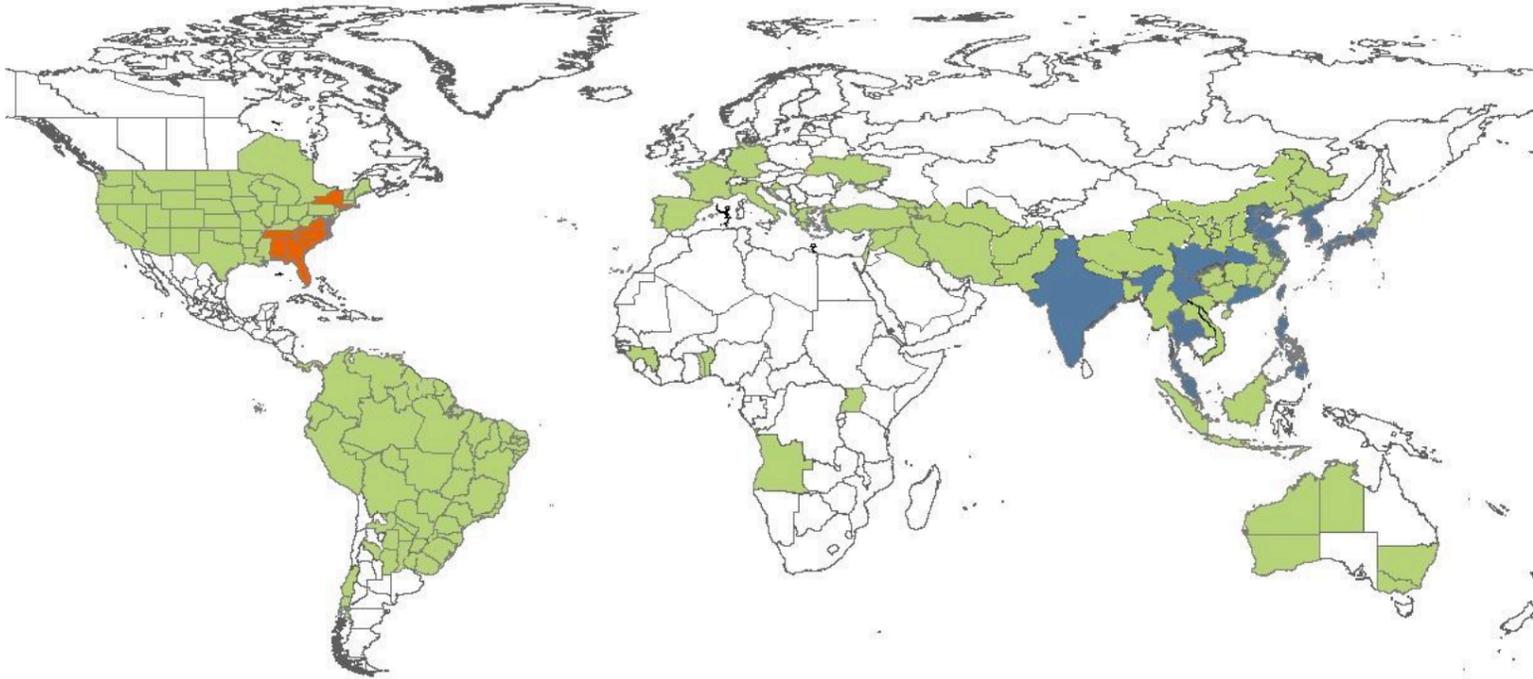


Figure 1.3 Distribution of *Reticulitermes* and *Coptotermes* allopatric (green) and sympatric distributions with *Pachycondyla* (= *Brachyponera*) *chinensis* in native (dark blue) and introduced (orange) ranges.

### ***Pachycondyla chinensis* detection of *R. virginicus* olfactory cues**

*Pachycondyla* (= *Brachyponera*) *chinensis* chose the direction towards the Petri dish containing *R. virginicus* 80% of the time regardless of left/right orientation of termite placement (Pearson  $\chi^2 = 30$ ,  $df = 1$ ,  $p < 0.0001$ ), revealing that *P. chinensis* detected *R. virginicus* from a distance of at least 7cm. This confirmed our observations during the prey acceptance study, where *P. chinensis* foragers appeared to ‘track’ *R. virginicus* workers. However, when presented with a choice between *R. virginicus* workers and *Parcoblatta* nymphs ( $\chi^2 = 1.8$ ,  $df = 1$ ,  $p = 0.17$ ) or collembolans ( $\chi^2 = 0.05$ ,  $df = 1$ ,  $p = 0.81$ ) *P. chinensis* did not choose the direction of the termites more frequently. Similar results were obtained when *P. chinensis* workers were exposed to cellulose powder fed on by termites compared to that which had not ( $\chi^2 = 0.8$ ,  $df = 1$ ,  $p = 0.37$ ).

### ***Pachycondyla chinensis* nest choice assays**

#### Lab Assay:

Most *P. chinensis* workers vacated the plaster nest and moved into the cellulose matrix containing *R. virginicus* within the first 5 hours of introduction to the arena. We continued recording their location during the next 5 days to see if the ants would continue inhabiting the termite nest. *Pachycondyla chinensis* were recorded in the termite nest during the entire 5-day period. We were able to reject the null hypothesis that the frequency of *P. chinensis* workers would be found at equal rates between the three locations ( $\chi^2 = 10.95$ ,  $df = 2$ ,  $p < 0.05$ ). We observed *P. chinensis* workers in the nest containing termites three times more often than the one without (figure 1.4). We observed worker *P. chinensis* carrying other

workers in their mandibles from their original nest into the cellulose matrix occupied by *R. virginicus* as well as into the unoccupied cellulose matrix.

Field assay 1:

Bait stations that were neither inoculated with *R. virginicus* nor moistened never contained *P. chinensis* or *R. virginicus* during the 3-month pretrial period.

Field assay 2:

Seventy-five percent of the stations inoculated with *R. virginicus* were occupied by *P. chinensis*, whereas zero percent of stations lacking termites contained *P. chinensis* ( $\text{Chi}^2=41.311$ ,  $p<0.0001$ ). After six days, 49% of the stations that had been inoculated with termites no longer contained termites, leading to a 57% decrease in stations occupied by *P. chinensis* between the 3<sup>rd</sup> and 6<sup>th</sup> day.

**Foraging strategies of *P. chinensis* workers in the presence and absence of termites**

The first *P. chinensis* worker that encountered the termite nest entrance behaved in an excited manner while circling the entrance several times. When a termite worker came to the entrance, presumably to extend the tunnel, it was seized and stung. The termite was released after it became immobilized, whereupon the ant again circled the nest. After ca. 2 minutes the worker collected the dead termite and returned it to the *P. chinensis* nest. Once inside the ant nest many workers pulled at the termite and began feeding upon it. Approximately five workers then rapidly evacuated the nest. The first worker that brought back the termite did not lead or carry

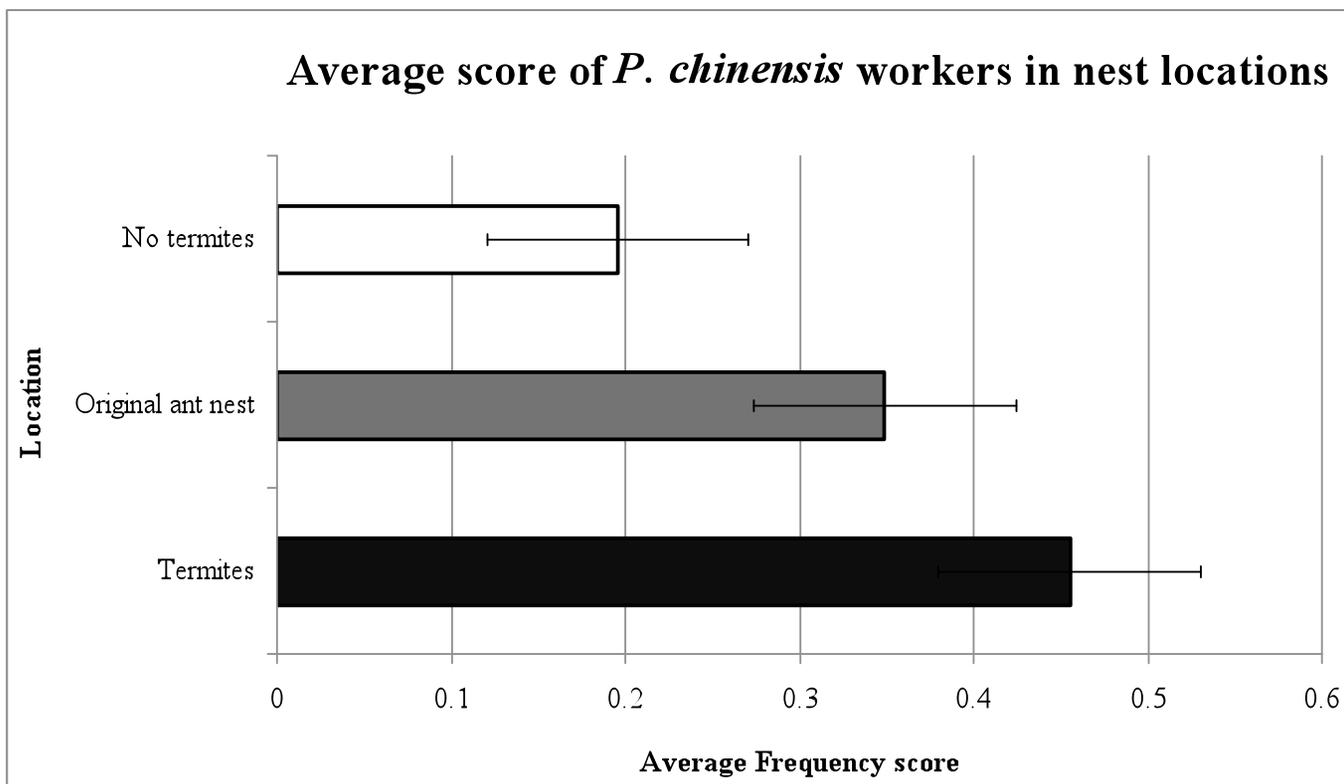


Figure 1.4 Average score of observations where *P. chinensis* occupied stations with termites (dark), without (open), and the original Petri dish nest (grey)

any workers back to the termite nest. *Pachycondyla* (= *Brachyponera*) *chinensis* workers that stung and killed *R. virginicus* at the nest entrance did not carry them back to the nest immediately. Frequently, other *P. chinensis* carried the dead termites back to the ant nest. After ca. 25 minutes ca. 10 workers were at the *R. virginicus* nest entrance, removing termites as they appeared.

Despite no active recruitment, there was a dramatic increase in foragers at the termite nest entrance within the first hour. Workers contacting the termite nest entrance remained three times higher, on average, than the control replicates during the 24 hours of observation ( $F=6.73$ ,  $df=1,8$ ,  $p=0.0319$ , Figure 1.5a). When other workers contacted the termite nest entrance their behavior was similar to the first; including excited circling, and the occasional removal of foraging termites. Interestingly, *P. chinensis* workers did not enter the termite nest at a higher rate even when termites were present ( $F=1.04$ ,  $df=1,8$ ,  $p=0.3379$ , Figure 1.5b). We show that even in the presence of termites ants are six times more likely to contact the nest entrance than they are to enter it over a 24-hour period ( $F=10.5$ ,  $df=1,8$ ,  $p=0.0118$ ).

## Discussion

We provide five lines of evidence that subterranean termites are an important resource for *P. chinensis* and aid in their successful introduction and spread throughout new habitats. First, *P. chinensis* do not occur in habitats lacking Rhinotermitidae. Second, of the potential prey near *P. chinensis* colonies, *R. virginicus* workers were preferred overwhelmingly. Third, *P. chinensis* oriented towards *R. virginicus* from a distance in an olfactometer, suggesting the ability to

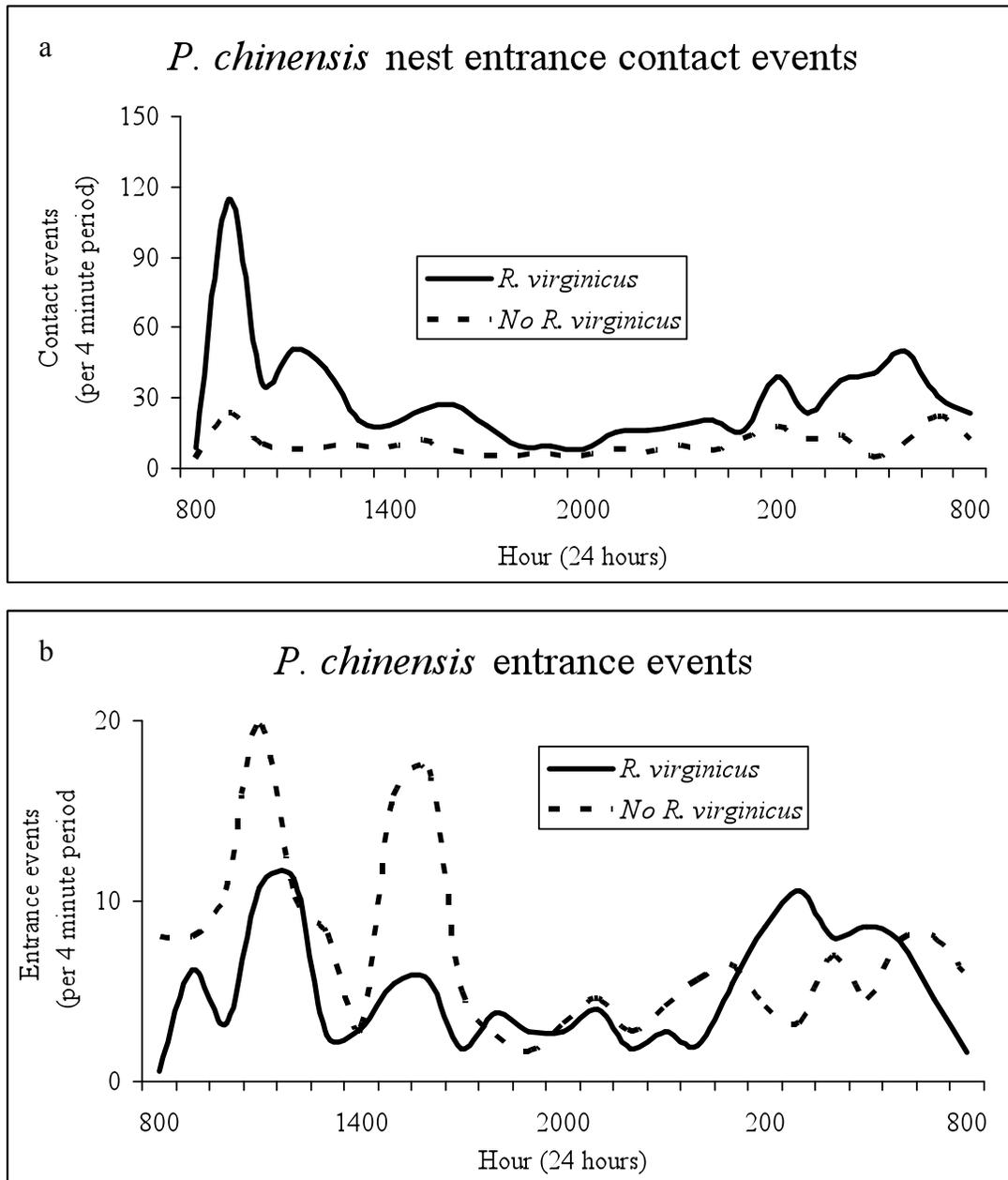


Figure 1.5 a) Number of *P. chinensis* workers that contacted the termite nest entrance in the presence (solid line) and absence (dashed line) of *R. virginicus*. b) Number of *P. chinensis* workers entering the termite nest in the presence (solid line) and absence (dashed line) of *R. virginicus*.

detect termite odors as well as other prey items. Fourth, *P. chinensis* workers occupied *R. virginicus* nests in the lab and field. Finally, *P. chinensis* workers elicit behaviors enabling them to capture *R. virginicus* individuals. We conclude that the behavior of *P. chinensis* in the presence of termites increases their competitive abilities in natural areas where they have been introduced.

The sympatric distribution of *P. chinensis* with wood feeding termites (Rhinotermitidae) suggests that termites are critical for *P. chinensis* survival. Rhinotermitidae have a pantropical-temperate distribution and include some of the most serious pest species (e.g. *Coptotermes formosanus*) (Eggleton 2000). The geographic distribution of *Pachycondyla marginata*, a termite specialist feeding on *Neocapritermes opacus*, largely overlaps with its prey, as do other obligate termitophages (Leal and Oliveira 1995) (See Table 1.2 for a more complete list).

Many different arthropods occurring in and around logs occupied by *P. chinensis* and considered as potential prey were largely ignored in our laboratory assay. It is possible that during our field surveys we missed collecting some important arthropod prey; however, if these prey were indeed an important part of the diet of a large invasive ant colony they should be fairly conspicuous. It is possible that the response of *P. chinensis* workers to the prey that we did offer might have been constrained in our laboratory assay; however, *R. virginicus* workers were rapidly located, attacked and retrieved. Many invasive ant species decimate local arthropod fauna (Holway et al. 2002) and subsequently switch their diet to that largely comprised of hemipteran honeydew (Abbott and Green 2007, Tillberg et al. 2007); an

**Table 1.2 Distributions of termitophagous ants and their prey.**

Obligate Termitophages	Termite prey	Ant distribution	Termite distribution	Reference
<i>Pachycondyla marginata</i>	<i>Neocapritermes opacus</i>			(Leal and Oliveira 1995)
<i>Pachycondyla laevigata</i>	<i>Neocapritermes, Microcerotermes, Coptotermes, Armitermes</i>	Neotropical	Neotropical	(Hölldobler and Traniello 1980)
<i>Pachycondyla commutata</i>	<i>Syntermes</i>			(Mill 1984)
<i>Cylindromyrmex striatus</i>	<i>Nasutitermes</i>			(Overall and Bandeira 1985)
<i>Pachycondyla analis</i> (= <i>Megaponera foetens</i> )	<i>Macrotermitinae</i>			(Lepage 1981, Corbara and Dejean 2000)
<i>Pachycondyla caffraria</i>	<i>Microcentrotermes</i>	Afrotropical	Palaeotropical (includes Afrotropics and Indomalayan zones)	(Agbogba 1992)
<i>Decamorium uelense</i>	<i>Macrotermitinae</i>			(Longhurst et al. 1979)
<i>Centromyrmex bequaerti</i>	<i>Macrotermitinae</i>			(Dejean and Feneron 1999)
<i>Leptogenys chinensis</i>	<i>Macrotermitinae</i>	Indomalayan		(Maschwitz and Schonegge 1983)

abundant, high quality, renewable food supply. We found no evidence that *P. chinensis* workers tended honeydew-excreting hemipterans at or above the soil surface. In fact, ponerine ants in general do not tend hemipterans because they do not possess a crop. The crop allows ant species to share liquid resources between nest mates through trophallaxis (Paul and Roces 2003). It is unlikely that hemipteran honeydew (comprised mostly of liquid simple carbohydrates) is a significant dietary component.

Yet, *R. virginicus* and other termites may provide benefits to *P. chinensis* invaders that are similar to the value of hemipteran honeydew for other invasive ants: they are abundant (Wood and Sands 1978), of high nutrient value (Sogbesan and Ugwumba 2008), easily processed (possessing thin cuticle), difficult to overexploit (refuge in narrow galleries and defensive soldier caste (Prestwich 1984, Jaffe et al. 1995, Matsuura 2002, Buczkowski and Bennett 2008) and renewable (numerous progeny produced year-round (Matsuura et al. 2009)). *Pachycondyla chinensis* workers exhibit plasticity in their feeding habits, however, it is likely that termites increase their ability to produce more workers and gynes as in other specialists (for example *Plectrotena minor* (Suzzoni et al. 2000)), thus providing a springboard for their success. The access to a high quality food source year round allows invasive ants, like *P. chinensis*, to exploit dietary nitrogen. Native ant species may also be reliant on termites as a source of nitrogen (Davidson 1997, Bluthgen et al. 2003, Palmer 2003, Dejean et al. 2007).

*Pachycondyla* (= *Brachyponera*) *chinensis* workers oriented to *R. virginicus* workers prior to physical contact, suggesting a role for olfactory cues in prey detection. However, the response to prey items is most likely general, i.e. to CO<sub>2</sub>. We determined, in our olfactometer

experiment, that *P. chinensis* workers detect odorants emitted by all prey items separate from its nesting material. While we determined that the ants moved into nests containing termites, it was interesting that *P. chinensis* workers did not respond differently to substrate previously occupied by termites.

We determined that *P. chinensis* workers moved into *R. virginicus* nests. Nests, both occupied and abandoned by termites, provide shelter for invertebrates (Dejean et al. 1996) including 75 different species of ants, as well as non-native termites (Dejean et al. 1997). Upon inspecting the stations occupied by *P. chinensis* workers in the lab and field, we found ants had excavated the cellulose matrix. Modification of termite tunnels by *P. chinensis* workers as they prey upon termites suggests a certain level of termitolesty.

*Eurhopalothrix heliscata* (Wilson and Brown 1984), *Pachycondyla caffraria* (Agbogba 1992), and *Centromyrmex bequaerti* (Dejean and Feneron 1999), exhibit varying degrees of termitolesty, where the ants live with and prey upon termites. Wilson and Brown (1984) suggested that by living close to their food source, *E. heliscata* might have a competitive advantage over other ant species. By occupying termite galleries, *P. chinensis* workers possess a competitive advantage over other ground-nesting ants that prey on termites, such as *Aphaenogaster rudis* (complex).

We recorded a 3-fold increase in *P. chinensis* workers foraging at the entrance to *R. virginicus* nests compared with unoccupied nests. This behavior had not been previously recorded for other *Pachycondyla* species that mass raid when hunting for termites. Following discovery of a termite nest, a scout *Pachycondyla analis* deposits a pheromone trail while returning to its nest and then recruits a raiding party, which it leads to the termite nest. Ant

workers enter the nest bringing termites back to the outside nest surface. (Wheeler 1936, Corbara and Dejean 2000, Bayliss and Fielding 2002) We did not observe mass nest raiding in *P. chinensis* workers. Instead, *P. chinensis* foragers amassed at the termite nest entrance and grabbed *R. virginicus* individuals as they appeared. Many *P. chinensis* workers at the termite nest entrance may interfere with native ant species ability to prey upon termites.

Subterranean termites are widespread and abundant in the Southeastern U.S. (Watt et al. 1997) and thus may constitute a critical food source for *P. chinensis*, facilitating its establishment and spread in deciduous and coniferous forests. While subterranean termites may have a positive near-term impact on *P. chinensis*, invasive *P. chinensis* may deplete native subterranean termite populations, with cascading effects at other trophic levels. Subterranean termites burrow through soil and wood, consuming cellulose, and thus contribute to litter decomposition, soil aeration, and carbon recycling (Wood and Sands 1978, Dejean et al. 2007). Other invasive ant species have been shown to reduce the number of termite colonies (Dejean et al. 2007). Therefore, this ant may negatively impact long-term ecosystem processes.

## CHAPTER 2

### **Ant wars: Competition for termite resources between the native ant *Aphaenogaster rudis* (complex) and the invasive ant, *Pachycondyla* (= *Brachyponera*) *chinensis***

Invasive species are often thought to possess abilities, which give them a competitive advantage over native species, ultimately leading to the demise of the latter. In ants competition is especially important in structuring community assemblages (Brian 1965a, b, c, d, Fellers 1987, Herbers 1989, Hölldobler and Wilson 1990, Amarasekare 2002, Sanders and Gordon 2003). Typically a trade-off between interference and exploitation competition in the dominance and discovery of resources acts in a balanced manner allowing several ant species to coexist (Fellers 1987, Davidson 1998). The introduction of novel ant species often disrupts the balance of a community structured by competition (Case and Gilpin 1974, Mooney and Cleland 2001, Reitz and Trumble 2002), especially when trade-offs are broken (Davidson 1998, Amarasekare 2002). Here, we examine the direct and indirect interactions between the invasive ant, *P. chinensis*, and a native ant, *A. rudis* (complex), as they compete for a shared resource, *R. virginicus* termites.

In ants, direct/interference competition involves aggressive interactions between those at the individual and group level. An ant with less combative individuals may be more prone to engage in aggressive behavior if nest mates are nearby to assist her. Fighting ability can increase with numerical dominance changing the dynamics of interactions (Holway 1999). Fighting ability in ants is not limited by size or potency of venom between species; for example, the Argentine ant, a small stingless ant, raids harvester ant nests which are much larger and possess a powerful sting (Zee and Holway 2006).

Indirect/exploitative competition occurs when two species are competing for the same resource. The species that uses the resource most efficiently will displace the other (Gause 1934, Gause et al. 1934, Hardin 1960). Ants increase resource use through numerical dominance, allowing for increased recruitment to remove a resource (Holway 1999), dispersed central place foraging (Holway and Case 2000), or resource specialization (King and Tschinkel 2008). Often a combination of interference and exploitative competition by invasive ants leads to the displacement of natives (Holway et al. 2002, Reitz and Trumble 2002).

The Asian needle ant, *Pachycondyla* (= *Brachyponera*) *chinensis*, is native to tropical regions of Australasia (Wheeler 1930), but was introduced into the Southeastern United States in the 19<sup>th</sup> century (Smith 1934). Since its introduction to the U.S., *P. chinensis*, has been shown to cause an overall decrease in ant species diversity, most notably displacing *Aphaenogaster rudis* (complex) from woodland habitats, making this ant an invasive (Guénard unpublished data). *P. chinensis* and *A. rudis* (complex) share a similar niche involving termites. Both ants rely on termites for food and nesting substrate (Smallwood 1982, Southerland 1988, Buczkowski and Bennett 2007). It is not known at what levels interference and exploitative competitive interactions play a role in the displacement of *A. rudis* (complex) by invasive *P. chinensis* ants.

We hypothesize that the displacement of *A. rudis* (complex) by *P. chinensis* occurs through both direct (aggression and/or predation) and indirect (consumption of *Reticulitermes* spp.) competition. We examined direct behavioral interactions between *P. chinensis* and *A. rudis* (complex) ants in one-on-one (Tanner and Adler 2009) and colony

level (Morrison 2000) assays. We studied indirect effects between *P. chinensis* and *A. rudis* (complex) by assessing worker foraging ability by comparing each ants' impact on *Reticulitermes virginicus* mortality. We hypothesized that *P. chinensis* more efficiently consumed *R. virginicus*, thereby leaving less of this resource for *A. rudis* (complex).

## **Methods and Materials**

### **Collection of colonies**

We collected *P. chinensis*, *A. rudis* (complex), and *R. virginicus* nests from downed rotting logs at Schenck Memorial Forest (N 35.81737 and W -78.72778. Schenck Forest is dominated by 10 – 70 yr *Pinus taeda*. We aspirated 50 *A. rudis* (complex) colonies while in the field and placed each colony into separate containers. We counted the numbers of individuals within each *A. rudis* (complex) colony, a minimum of 75 workers, one queen, and several brood were placed into fluon-coated containers (Ziploc, 591ml), if fewer were captured they were returned to the collection site. A test tube (15mL) one-half full with water and plugged with cotton served as a nest. Five logs containing a single *P. chinensis* nest each and *R. virginicus* termites were returned to the lab and extracted from wood debris. We were not sure if nests from the five logs contained ants from the same colony, but each was treated as a separate colony. We housed each *P. chinensis* colony in a fluon-coated bin (76x76x6.4cm) with four moist plaster nests (140x15mm). We sorted wood fragments containing *R. virginicus* into five bins containing four Petri dishes (140x15mm) filled with a cellulose matrix.

We fed ant colonies 10 *R. virginicus* daily and provided 1.5mL artificial diet, *ad libitum* unless otherwise stated. Ant diet consisted of casein, sugar, salt, a crushed multi-

vitamin, potassium phosphate, magnesium chloride, cholesterol, water, and Kappa Carrageenan (Bhatkar and Whitcomb 1970). All colonies were maintained at constant temperature and humidity (26.7°C, 80%) on a 12:12 L/D cycle.

*Reticulitermes virginicus* were identified using molecular techniques (see chapter 1).

### **Direct Effects**

#### **Do single *P. chinensis* workers defeat *A. rudis* (complex) workers?**

We determined which species, *P. chinensis* or *A. rudis* (complex), had more aggressive individuals in one-on-one encounters with each other. We placed ants into an arena similar to the one described by Buczkowski and Bennet (2008). We picked up ants with a moist cotton swab and placed them into a fluon-coated beaker (50mL). The resident ant was given a one-minute acclimation period before the intruder was added. To account for intruder effects, *P. chinensis* workers were placed in the arena first in one-half of the replicates, while *A. rudis* (complex) workers were added first for the remaining replicates.

We recorded the:

- 1) number of encounters (distinct physical contact between individuals),
- 2) maximum aggression score for all encounters (0=no discovery, 1=ignore, 2=avoid, 3=biting/lunging, 4=stinging),
- 3) initiator of the aggressive interaction,
- 4) time until death since the 1<sup>st</sup> interaction, and
- 5) winning member, if any.

We analyzed the proportion of instances across replicates for each interaction grouped by intruder species. Data were then pooled to compare overall mortality in each ant species and analyzed using  $\chi^2$  with JMP (SAS-v7.0 2007).

**Do interactions between *P. chinensis* and *A. rudis* (complex) colonies change in the presence of *R. virginicus* prey?**

We tested for interference and exploitation competition between *A. rudis* (complex) and *P. chinensis* in the presence of *R. virginicus*. We connected three trays (25 cm x15 cm x11cm) in succession to one another using nalgene tubing (8cm x 8mmID) (Figure 2.1). We placed a moistened wooded block (15 cm x4 cm x2cm) with empty termite galleries, 75mL of moistened (1:1;V:V) cellulose and 1.5mL artificial ant diet in each tray.

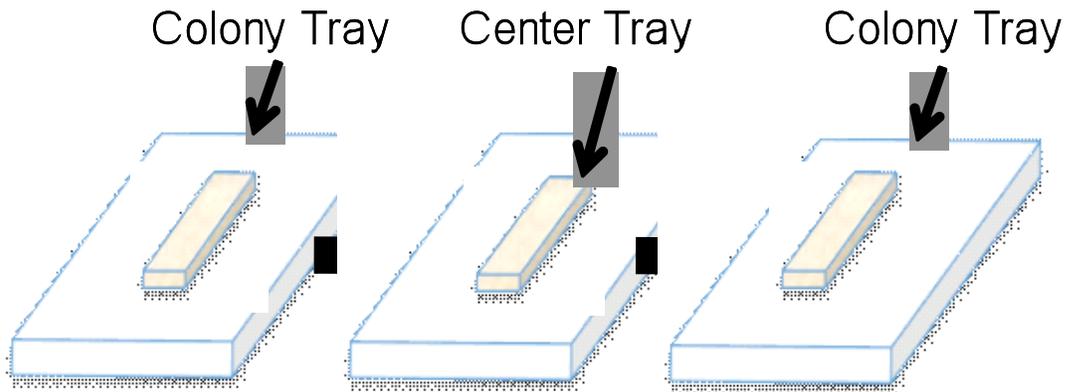


Figure 2.1 Arrangement for laboratory experiment testing the effects of termite presence on interactions between *P. chinensis* and *A. rudis* (complex).

The center tray was provisioned with *R. virginicus* (ca. 1000, 1% soldiers) in one-half of the replicates. Each outside tray (ant colony) in a series contained either an *A. rudis* (complex) or *P. chinensis* colony. We blocked ant access to the foraging arena for 24 h prior to the beginning of the experiment, after which time ants and termites could interact for three

days. Heterospecific ant colonies were of identical size for each replicate. All colonies contained an average of  $150 \pm 62.6$  workers including 1 queen (ca. 10 brood were included, but not counted as part of the total). Ten replicates per treatment were performed.

We recorded which ant species arrived first in the center tray. We also recorded the number of each ant species in the center tray and ant colony trays, every 20 minutes, during the first 80 minutes. Where both *P. chinensis* and *A. rudis* (complex) occurred in the same tray we recorded the number, nature and location of interactions for 5 minutes. We classified three types of interactions as either attack, retreat/avoid, or coexist. These recordings were made on each of three days. We also removed and recorded the number of all dead ants daily. We recorded the location and number of all live remaining ants at the end of the experiment. In order to observe the frequency of movement between trays by *P. chinensis* video cameras recorded activity at the entrance to the *A. rudis* (complex) nest during the entire trial. This allowed us to determine whether *P. chinensis* raided *A. rudis* (complex) nests (Zee and Holway 2006). We compared the number of live ants remaining at the end of the experiment between treatments using an ANOVA, and the type of interaction in each location was analyzed using a  $\text{Chi}^2$  analysis in JMP (SAS-v7.0 2007).

### **Indirect Effects**

**Are *P. chinensis* workers more effective at killing individual *R. virginicus* workers and soldiers than *A. rudis* workers?**

We determined whether individual *P. chinensis* workers were more effective than *A. rudis* workers in killing *R. virginicus* workers and soldiers. We placed insects into an arena similar to the one used by Buczkowski and Bennett (2008). We collected termites with a

moist cotton swab and placed them into a fluon-coated beaker (50mL). After 1 minute, either one *A. rudis* or one *P. chinensis* was placed into the container with the termite. We recorded the same behaviors as above with one-on-one *P. chinensis*-*A. rudis* (complex) interactions. Twenty-five replicates were performed for each ant species/*R. virginicus* pairing. We compared the time (seconds) and the number of encounters to kill the termites, for each ant species with one-way analysis of variance. We compared the proportion of termites killed for each ant species using a Chi<sup>2</sup> analysis in JMP (SAS-v7.0 2007).

### **Does termite soldier ratio affect *P. chinensis* and *A. rudis* (complex) ability to kill termites?**

Soldier ratio has not been shown to have an effect on ant predation. However, different ant species may differ in their ability to capture and consume termites (Cornelius and Grace 1995, 1996, 1997). We varied the number of *R. virginicus* soldiers and measured the total number of termites killed by either *P. chinensis* or *A. rudis* (complex).

We exposed *R. virginicus* to either *P. chinensis* or *A. rudis* (complex) at a 2:1 ratio. Termite soldiers represented 1, 5, or 10% of the total population evaluated. We are not aware of actual *R. virginicus* soldier:worker field ratios, but nests of a related species, *R. flavipes*, has been reported to contain 2.6% soldiers (Long et al. 2003, 2007).

We placed termites (200) in Petri dishes (60 mm dia x15mm) containing sawdust for 24 h, during which time they constructed carton tunnels extending from an entrance consisting of plastic tubing (10 mm x 2 mm ID). Ants (n = 100) were housed in moist plaster filled Petri dish (100 mm x15 mm) connected to an empty Petri dish by 10 mm of tubing, but was blocked for 24-hours, thus preventing access to the termites (Figure 1.2). We provided

the ants with a 25% sucrose solution and freshly killed cockroaches, *Blattella germanica*, *ad libitum* throughout the experiment. Access was subsequently allowed, whereby termites and ants could interact. After 7 days, we counted the number of surviving termites. Control (no ant) mortality of *R. virginicus* was also measured at each soldier ratio.

Adjustments for control mortality were made with the Henderson-Tilton formula (1955) where the corrected proportion of *R. virginicus* killed by ants is:

Proportion killed by ants =  $(1 - (C_b * T_a) / (C_a * T_b))$ ; Where:

- $C_a$  = Control Insect population after
- $C_b$  = Control Insect population before
- $T_a$  = Treated Insect population after
- $T_b$  = Treated Insect population before

We analyzed these data using a one-way analysis of variance between the two ants at each soldier ratio, with multiple comparisons (Tukey-Kramer HSD) in JMP (SAS-v7.0 2007).

### **Do termite-constructed barriers affect predation by *P. chinensis* and *A. rudis* nests differentially?**

We compared predation by *P. chinensis* and *A. rudis* (complex) nest fragments on *R. virginicus* nest fragments within solid wood or sand nests. Natural wood barriers are much more substantial and have been shown to prevent *A. rudis* (complex) workers from causing high *R. flavipes* mortality (Matsuura 2002, Buczkowski and Bennett 2008). *Reticulitermes* *sp.* use phragmosis, the plugging of entrances with soldier heads, for nest defense. Soldier presence using phragmosis has prevented high mortality of *R. speratus* against *P. chinensis*

(Matsuura 2002). We predicted that phragmosis was more effective in tunnels within solid wood.

*Reticulitermes virginicus* workers (ca. 10,000) were placed in a container (189L) with moist soil and pine wood blocks (15 cm x 4 cm x 2cm) for 60 days. After galleries were constructed we removed the termites from the blocks as described in Buzckowski and Bennett (2008). Briefly, wooden blocks containing termites were placed on a moist piece of corrugated cardboard under an intense fluorescent light. As the wooden block dried the termites moved into the moistened cardboard. We then re-moistened the wooden blocks by soaking them in distilled water for 24 hr. We compared the effect of ant predation on termites within these wooden block nests with that of nests constructed within moist soil-filled Petri dishes (100 mm dia x 15 mm) provisioned with moist filter paper (78 cm<sup>2</sup>). A wooden block or Petri dish was placed in a plastic fluon-coated container (26 cm x 17 cm x 4 cm) filled with moist sand (442 cm<sup>2</sup>) and 200 *R. virginicus* (5% soldiers). These containers were connected to a container of the same size by nalgene tubing (8 cm x 8 mm ID), which housed either *P. chinensis* (100 workers, 1 queen, and 50 brood), or *A. rudis* (complex) (100 workers, 1 queen, and 50 brood). Containers housing ants were provisioned with a wooden block nest, and food (20% sucrose solution and 10 live *R. virginicus* workers) (Figure 2.1 minus second colony tray).

Treatments lacking ants were included to measure *R. virginicus* natural mortality. Ants were first isolated from termites for 24 h by blocking the connecting tube. Then the tube was unblocked, thus allowing movement between the containers. We performed 13 replicates

for each pair-wise interaction. After seven days, we counted all living termites, after forcing them from the wooden blocks by drying.

We calculated termite mortality by subtracting the remaining termites at the end of 7 days from 200 and used the Henderson-Tilton (1955) correction for untreated control (no ant) mortality. We compared termite mortality using a one-way analysis of variance between the two ants for each termite barrier treatment, with multiple comparisons (Tukey-Kramer HSD) in JMP (SAS-v7.0 2007).

**Are *P. chinensis* workers more effective at capturing termites than *A. rudis* (complex) workers?**

We observed whether *A. rudis* (complex) and *P. chinensis* use the same foraging strategy when hunting for termites by recording their behavior in the presence and absence of termites.

Two hundred *R. virginicus* individuals (198 workers plus 2 soldiers) were housed in a Petri dish (60x15mm) nest containing moist pine wood sawdust. This nest was connected to an empty Petri dish (50x9mm) by plastic tubing (10mm x 2mmID). Individuals of *Pachycondyla* (= *Brachyponera*) *chinensis* or *A. rudis* (complex) (100 workers, 1 queen, and 10 brood) were housed in a moist plaster-filled petri dish (100x15mm), which was attached to the above empty petri dish (60x15mm) on one end and another empty Petri dish distal to the termite nest (Figure 2.4). This dish was provisioned with 25% sucrose solution and freshly killed cockroaches, *Blattella germanica*, *ad libitum*. Interactions between the ants and *R. virginicus* were inhibited for 24 hours prior to the beginning of recording by blocking the tube entering the dish with the termites.

We recorded the behavior of *P. chinensis* and *A. rudis* (complex) in response to sawdust-filled nests with and without *R. virginicus*. Observations of the ant's contact with or entry into the termite (or empty) nest entrance were recorded for four minutes every hour for 24 hours. We considered that an ant worker entered when the entire length of its body was within the nest. If an ant foraged at the nest entrance and immediately entered the nest we only recorded this as an entering event. The number of ants foraging was an instantaneous count we made of all the ants present outside the ant and termite nests. We divided the number of foraging events by the number of foraging ants to create a proportion of possible termite encounters per foraging ant.

We used repeated measures analysis of variance to compare the number of foraging events per foraging ant in the treatments between ants over time in JMP (SAS-v7.0 2007).

## **Results**

### **Direct Effects**

#### **Single *P. chinensis* workers defeat *A. rudis* (complex) workers**

*Pachycondyla chinensis* workers typically won in direct interactions with *A. rudis* workers, an outcome that did not depend on which ant was the resident and which was the intruder. When pooled across intruder and resident categories, *P. chinensis* killed *A. rudis* (complex) in 40% of the interactions compared to 2.8% *P. chinensis* mortality ( $X^2=16.05$ ,  $df=4$ ,  $p=0.003$ ).

#### **Interactions between *P. chinensis* and *A. rudis* (complex) nests change in the presence of *R. virginicus* prey**

*Pachycondyla chinensis* was the dominant ant even more often when *R. virginicus* were present (100%) than absent (86%) ( $X^2=4.308$ ,  $df=1$ ,  $p=0.0379$ ) perhaps as a result of interaction frequency. When *R. virginicus* were present the number of interactions between *P. chinensis* and *A. rudis* (complex) foragers increased ( $X^2=4.791$ ,  $df=1$ ,  $p=0.0286$ ) within the *A. rudis* (complex) colony tray (figure 2.2a), but had no effect on the number of *A. rudis* (complex) ( $F=0.139$ ,  $df=6,14$ ,  $p=0.988$ ) or *P. chinensis* ( $F=1.51$ ,  $df=6,14$ ,  $p=0.244$ ) foraging in the termite arena.

More individuals of *A. rudis* (complex) than *P. chinensis* died during the experiment ( $F=5.64$ ,  $df=3,16$ ,  $p=0.0078$ ), which was not influenced by the presence of *R. virginicus* in the central nest ( $F=0.3$ ,  $df=3,6$ ,  $p=0.824$ ) (Fig. 2.2b). *Pachycondyla chinensis* stored the bodies of the dead *A. rudis* (complex) in their nests indicating that *P. chinensis* prey on *A. rudis* (complex).

### **Indirect Effects**

#### ***Pachycondyla chinensis* workers killed individual *R. virginicus* workers and soldiers more quickly than did *A. rudis* workers**

Single *P. chinensis* killed *R. virginicus* workers more than 8 times faster than *A. rudis* and 15 times more often (100% vs. 6.7%). *Pachycondyla chinensis* killed *R. virginicus* soldiers ca. three times faster than *A. rudis* (complex) workers and five times more often (83% vs. 16.7%). (Table 2.1)

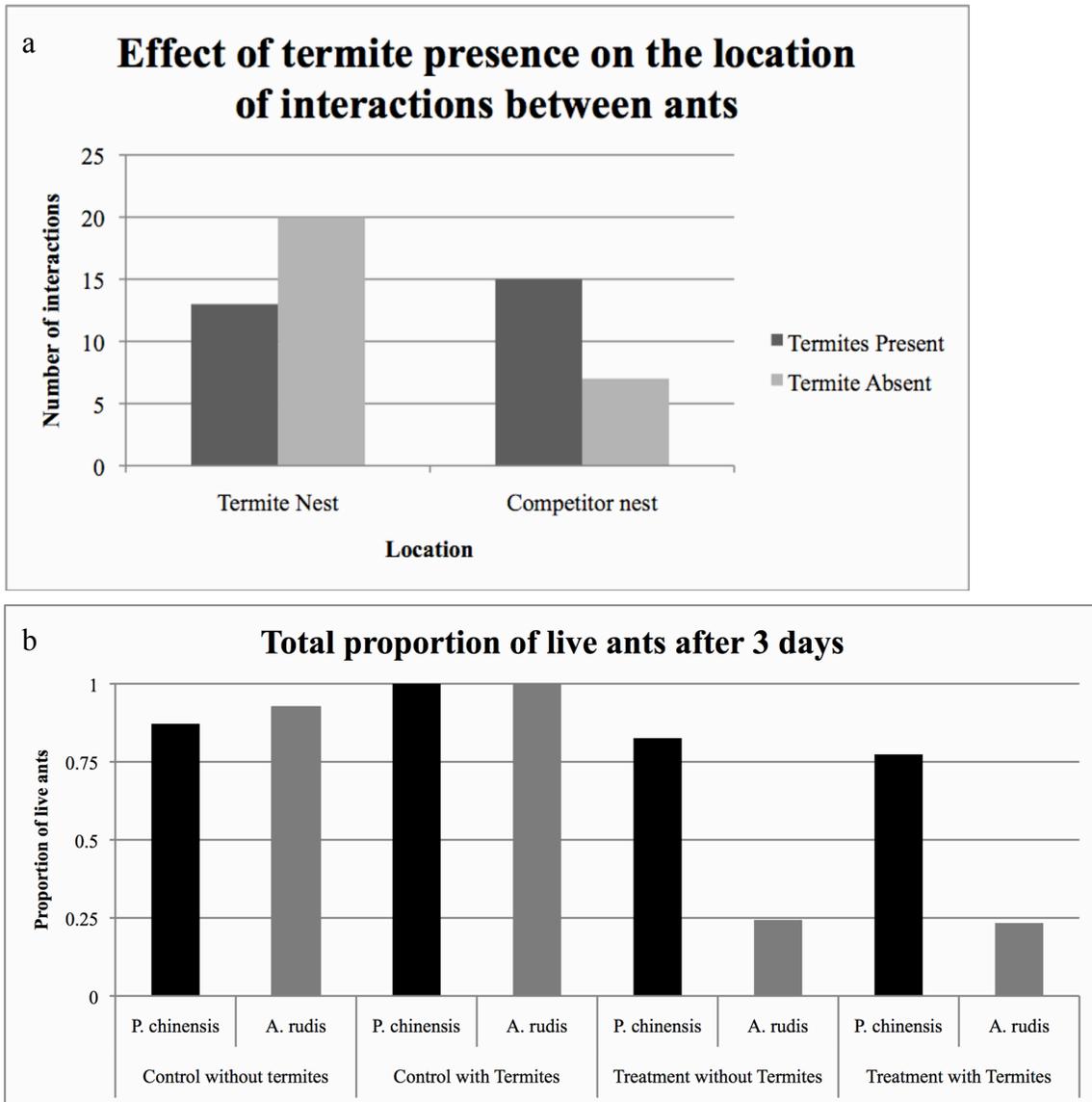


Figure 2.2 a) The number of encounters between *A. rudis* (complex) and *P. chinensis* in the presence (dark columns) and absence (light columns) of termites in the termite nesting arena (left) and competitor nesting arena (right). b) Mortality of *A. rudis* (complex) (light columns) and *P. chinensis* (dark columns) in the presence and absence of termites in competition (treatments) with the heterospecific ant and without (controls).

**Table 2.1 Results of one-on-one interactions between *P. chinensis*/*A. rudis* and *R. virginicus* \*For Tukey-Kramer HSD levels not connected by the same letter are significantly different at alpha=0.05.**

Resident	Intruder	Mean time to kill since 1 <sup>st</sup> interaction (seconds)	Percentage of time Termite killed
		Tukey-Kramer HSD* q=2.6, df=3	Pearson X <sup>2</sup> =84.25, df=9, p<0.0001
Termite Worker	<i>Aphaenogaster</i>	120 <sup>A</sup>	6.7%
	<i>Pachycondyla</i>	14.3 <sup>B</sup>	100%
Termite Soldier	<i>Aphaenogaster</i>	81.8 <sup>A</sup>	16.7%
	<i>Pachycondyla</i>	29.5 <sup>B</sup>	83.3%

### **Termite soldier ratio does not hinder *P. chinensis* ability to kill termites**

Similar to previous studies (Cornelius and Grace 1997) mortality caused by each ant species was not affected by different soldier ratios (F=0.079, df=2, p=0.924). However, when pooled across all ratios *P. chinensis* workers caused almost 6 times greater termite mortality than *A. rudis* (complex) (F=420.9, df=1, p<0.0001, Figure 2.3)

### **Termite predation by *P. chinensis* and *A. rudis* nests did not appear to be hindered by gallery walls of different materials**

All *Reticulitermes virginicus* in some of the initial 13 replicates were noted dead and covered with fungal mycelia. These replicates were not included in the analysis.

*P. chinensis* caused greater *Reticulitermes virginicus* mortality than *A. rudis* (complex) (F=11.98, df=5, p=0.0001). The nests with wooden galleries afforded no greater protection than sand nests against *P. chinensis* (F=3.46, df=2, p=0.0812) and *A. rudis* (complex) foragers (F=0.3517, df=2, p=0.5663, Figure 2.4).

### ***P. chinensis* workers forage for termites more efficiently than *A. rudis* (complex) workers**

The proportion of ant encounters at the termite nest entrance per foraging ant was five times higher for *P. chinensis* than *A. rudis* (complex) when termites were present ( $F=28.13$ ,  $df=1,8$ ,  $p=0.0007$ ; figure 2.5). This is interesting because each ant dispersed approximately the same amount of foragers in the presence of termites ( $F=0.228$ ,  $df=1,8$ ,  $p=0.6456$ ), and encountered the termite nest at similar rates, ( $F=4.23$ ,  $df=1,8$ ,  $p=0.0736$ ). We did not record termite mortality, but the trend was similar to previous studies where *R. virginicus* nests suffered greater casualties in the presence of *P. chinensis* nests.

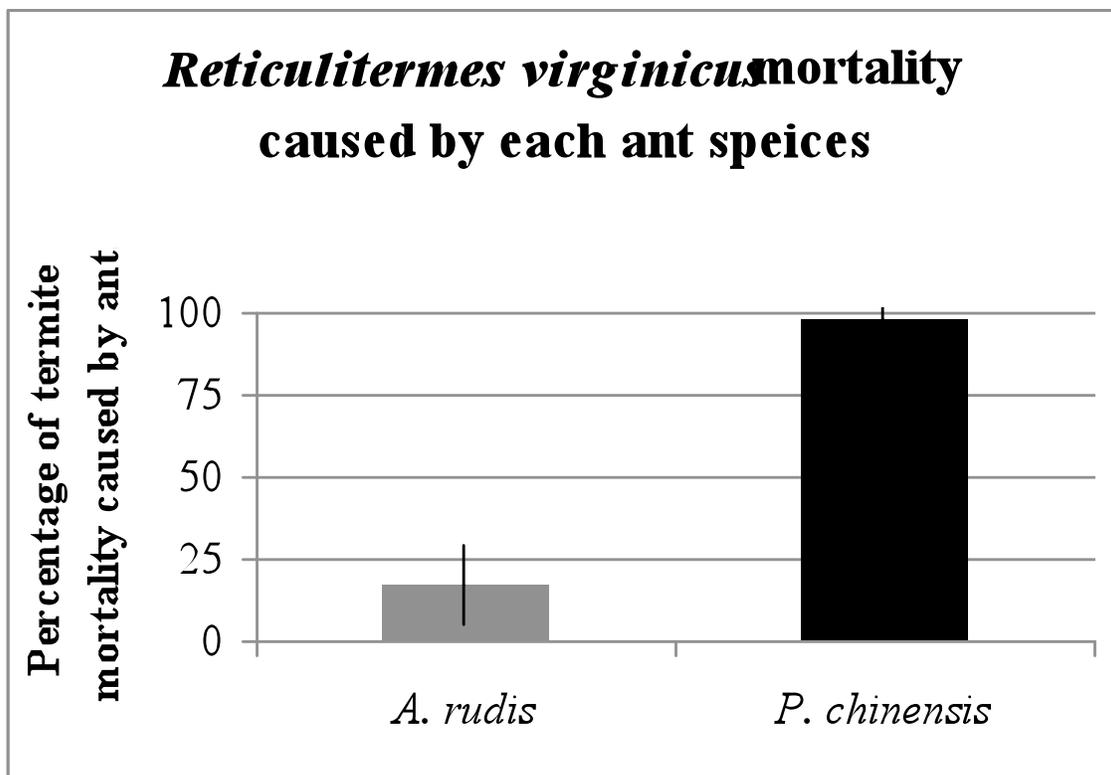


Figure 2.3 *Reticulitermes virginicus* mean ( $\pm$  standard deviation = 12.2) mortality, pooled across all soldier ratios, caused by *A. rudis* (lightly shaded) and *P. chinensis* (dark) workers (corrected for control mortality).

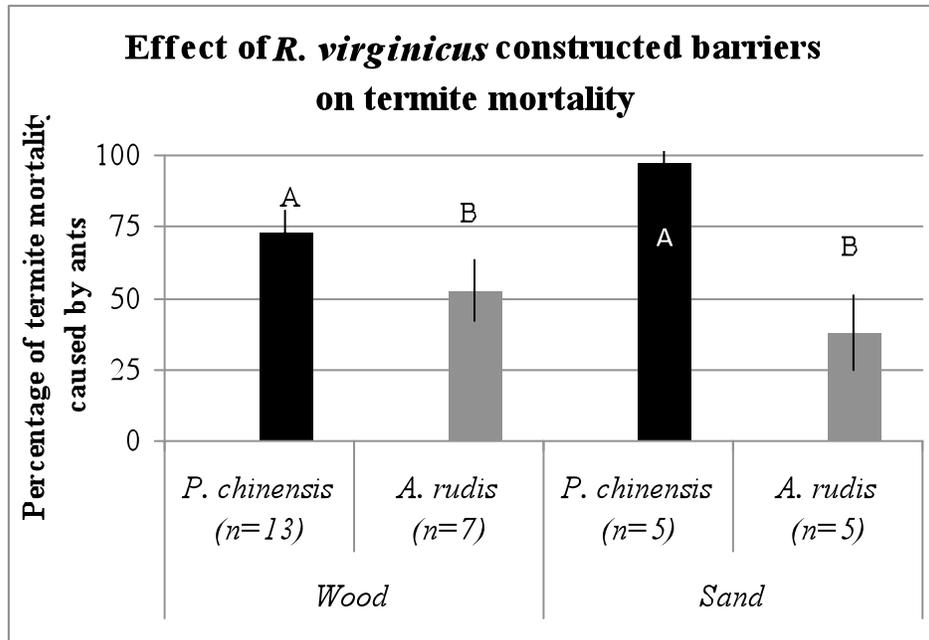


Figure 2.4 Average proportion of *R. virginicus* killed in nests with wood barriers (*Natural*) and sand nests (*Artificial*) by *P. chinensis* (dark) and *A. rudis* (light) workers corrected for control mortality. Bars with same letter are not significantly different ( $p < 0.05$ ).

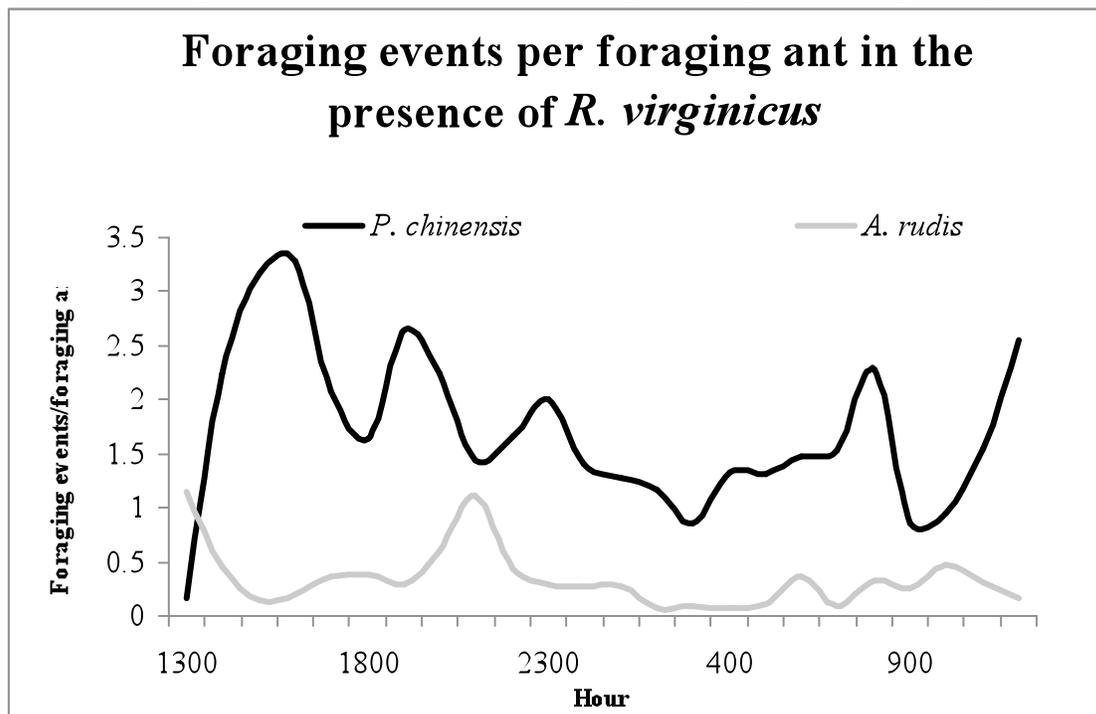


Figure 2.5. The number of foraging events per ant for *A. rudis* (light) and *P. chinensis* (dark) workers in the presence of termites over a 24 hour period.

## Discussion

*Aphaenogaster rudis* (complex) worker abundance is strongly negatively correlated with *P. chinensis* worker abundance (Guénard unpublished data). We provide support for two mechanisms that may, in part, be responsible for the displacement of *A. rudis* (complex) nests by *P. chinensis* colonies. First, *P. chinensis* dominate *A. rudis* (complex) workers in direct worker-worker and whole colony interactions, whereby most *A. rudis* (complex) are killed by *P. chinensis* workers. Second, both *A. rudis* (complex) (Buczowski and Bennett, 2007) and *P. chinensis* nests co-occur with and prey upon subterranean termites (*Reticulitermes spp.*); however, *P. chinensis* foragers discover nests, kill, and retrieve workers and soldiers of *R. virginicus* more quickly than *A. rudis* (complex) foragers. Therefore, termite prey may become limiting to *A. rudis* (complex) colonies.

### *Direct Effects*

Colony and individual level interactions between *P. chinensis* and *A. rudis* (complex) foragers reveal how domination of nest space may function in the displacement of *A. rudis* (complex) by *P. chinensis*. *Aphaenogaster rudis* (complex) largely remained within compartments containing their nests and thus direct heterospecific interactions were a consequence of the wider ranging exploratory activity of *P. chinensis* foragers. *Pachycondyla chinensis* subdued *A. rudis* (complex) workers in ca. 60 secs in most aggressive encounters. In the one instance where the *A. rudis* (complex) worker prevailed, the *P. chinensis* worker died ca. 240 seconds after being stung. Also, *R. virginicus* workers and soldiers died more quickly when stung by *P. chinensis* workers, compared to *A. rudis* (complex) workers. We suspect that *P. chinensis* worker

venom is more toxic than that of *A. rudis* (complex). Ponerines are known to possess some of the most potent venoms in the ant world (Piek et al. 1991), while Myrmicines are little studied for their venom (with the exception of *Solenopsis invicta* (Jones et al. 1982)).

Movement of *P. chinensis* foragers was greater in the presence of *R. virginicus*, which increased the number of interactions between *P. chinensis* and *A. rudis* (complex) workers in the *A. rudis* (complex) nest, producing higher ant mortality than when *R. virginicus* nests were absent. In Chapter 1, we demonstrated that a *P. chinensis* worker responded to volatile compound(s) produced by *R. virginicus* workers. These same compound(s) may have stimulated search patterns that increased encounter rates between *P. chinensis* and *A. rudis* (complex) foragers. *P. chinensis* colonies were partitioned between all available nests, sometimes abandoning the original nest, to live under the *R. virginicus* nest or within the *A. rudis* (complex) nest. Polydomy, dispersed central place foraging, and termitolesty have been shown to be an efficient strategy to secure resources (Wilson and Brown, 1984; Holway and Case, 2000; Schmolke, 2009).

Evidence from our individual and colony level assays suggests that a *P. chinensis* worker is practicing the "tit-for-tat" strategy. In game theory this strategy will always win because energy is only wasted if an aggressive action is forced to occur (Axelrod and Hamilton 1981) By cooperating and basing decisions on reciprocity of the conspecific the *P. chinensis* worker is able to fully utilize its time foraging with the minimum amount of energy wasted fighting. By contrast, *A. rudis* (complex) foragers must always be the aggressor in order to gain access to *R. virginicus* resources. While recruiting to the empty nest *P. chinensis* foragers also encountered and attacked *A. rudis* (complex) foragers at a higher rate

in the absence of termites, than in their presence. When termites were absent, interactions at the termite nest increased, presumably because *P. chinensis* workers were defending the nest space, as it was the first to discover and usually enter the nest.

#### *Indirect effects*

Exploitative competition structures ant communities (Fellers 1987, Offenberg 2001, Palmer 2003) and is a factor in the displacement of native by invasive ants (Dejean et al. 1993, Human and Gordon 1996, Holway 1999, Morrison 2000, Amarasekare 2002, Reitz and Trumble 2002, Lach 2005). Subterranean termites are prey for both *A. rudis* (complex) and *P. chinensis* colonies and superior prey capture efficiency by *P. chinensis* foragers may impact the abundance of *A. rudis* (complex) nests. More *P. chinensis* than *A. rudis* (complex) foragers were recorded at the entrance of *R. virginicus* nests, which may be an effective way to capture termites and minimize injury from termite soldier attack within the nest. These results are consistent with our findings reported in Chapter 1. The first *P. chinensis* worker that encountered the termite nest entrance behaved in an excited manner while circling the entrance several times. When a termite worker came to the entrance, presumably to extend the tunnel, it was seized and stung. Once inside the ant nest many workers pulled at the termite and began feeding upon it. Approximately 5 workers then rapidly evacuated the nest. This behavior could increase interactions with a competing ant like *A. rudis* (complex). We demonstrated that *P. chinensis* workers killed all *R. virginicus* individuals even when soldiers comprised 10% of the termite colony. A surreptitious strategy of capturing individual termites, including soldiers, at the nest entrance may explain the success of *P. chinensis* colonies, but not *A. rudis* (complex) foragers, in securing termite prey. However,

Buczowski and Bennett (2008) reported high predation rates of *A. rudis* (complex) on *R. flavipes*, even with 5% soldiers. Perhaps *R. flavipes* nests are less able to resist attack by *A. rudis* (complex) workers than *R. virginicus* nests. Cornelius and Grace (1996) also found no effect of *Coptotermes formosanus* soldier ratio on predation by two ant species.

Though Buczowski and Bennett (2008) reported a benefit of *R. flavipes* nests within solid wood against *A. rudis* (complex) predation, we recorded no such benefit to *R. virginicus* nests against either *A. rudis* (complex) or *P. chinensis* predation. *Reticulitermes* sp. use phragmosis for nest defense against arthropod, primarily ant, predators (Matsuura 2002) implying a benefit of high soldier ratios within wooden galleries as a defense against ants. A termite capture strategy utilized by *P. chinensis* foragers, as described above, might allow ants to evade this termite defense.

We demonstrated that termite defense systems such as soldier ratio and wood barriers have little if any hindering effects on *P. chinensis* forager ability to prey on termites. Perhaps *R. virginicus* colonies are pre-adapted for defense against the native *A. rudis* (complex) worker, but not the non-native *P. chinensis* worker. When prey species adjust to a novel predator, mortality rates may be initially high as seen in the iguanas of the Galapagos Islands after the introduction of feral cats and dogs (Berger et al. 2007). Novel predator interactions can lead to behavioral (Maloney and Mclean 1995, Kiesecker and Blaustein 1997, Wildy and Blaustein 2001) and physiological (Berger et al. 2007) changes in the prey. However, Cornelius and Grace (1995, 1996, 1997) showed that different ant species cause different levels of mortality in termites and concluded that different species of ants vary in their ability

to prey on termites. *R. virginicus* colonies are adapted to living with *A. rudis* (complex) nests, and wood barriers may be an effective means of limiting predation by this ant.

Different strategies for termite prey capture range from *raide en masse* (Wheeler 1936) where scout ants are necessary/unnecessary (Longhurst et al. 1979), to varying levels of termitolesty (Dejean and Feneron 1999), group recruitment (Dejean et al. 1993), or solitary foraging (Wilson and Brown 1984a). *P. chinensis* employs a combination of termitolesty, solitary foraging, and group recruitment to prey on *R. virginicus* nests causing higher mortality than *A. rudis* (complex) colonies (Chapter 1). Foraging patterns of *A. rudis* (complex) workers have shown that they are often the first to find a resource, but defend it poorly (Fellers 1987). Acquisition of *R. virginicus* as a food source is dependent upon the varied predatory strategies of *A. rudis* (complex) and *P. chinensis* foragers, potentially leading to indirect competitive effects.

Invasion of woodland habitat by *P. chinensis* throughout the Southeastern United States has placed them into competition with the native ant *A. rudis* (complex). Not only do these native and invasive ants share habitat, but food sources as well. An overlap of ecological niches between competing species will eventually lead to the displacement of the inferior competitor (Gause et al. 1934, Hardin 1960), and most often occurs when an exotic species is introduced into a new habitat (Reitz and Trumble 2002). Exploitative competition has not been shown to be the sole mechanism for displacement of native ants by invasive species. A combination of interference and exploitative competition probably occur, (Case and Gilpin 1974, Fellers 1987, Human and Gordon 1996, Reitz and Trumble 2002).

## REFERENCES

- Abbott, K. L. and P. T. Green. 2007. Collapse of an ant-scale mutualism in a rainforest on Christmas Island. *Oikos* **116**:1238-1246.
- Abril, S., J. Oliveras, and C. Gomez. 2007. Foraging activity and dietary spectrum of the Argentine ant (Hymenoptera : Formicidae) in invaded natural areas of the northeast Iberian Peninsula. *Environmental Entomology* **36**:1166-1173.
- Achetasham, A. and M. S. Akhtar. 2008. Swarming behaviour and alate sex-ratio of *Heterotermes indicola* (Wasmann) (Isoptera : Rhinotermitidae). *Pakistan Journal of Zoology* **40**:75-82.
- Agbogba, C. 1992. Settlement in the Prey Termite Nest by the Ponerine Ant *Pachycondyla caffraria* (Smith), and Tandem Running Signal Analysis for the Following Ant. *Journal of Ethology* **10**:133.
- Amarasekare, P. 2002. Interference competition and species coexistence. *Proceedings of the Royal Society Biological Sciences Series B* **269**:2541.
- Ambrose, H., K. Ambrose, D. Emlen, and K. Bright. 2002. A handbook of biological investigation. sixth edition. Hunter Textbooks Inc., Winston-Salem, NC.
- Anderson, C., G. Theraulaz, and J. L. Deneubourg. 2002. Self-assemblages in insect societies. *Insectes Sociaux* **49**:99-110.
- Austin, J. W., A. L. Szalanski, R. Ghayourfar, A. Kence, and R. E. Gold. 2006. Phylogeny and genetic variation of *Reticulitermes* (Isoptera : Rhinotermitidae) from the eastern Mediterranean and Middle East. *Sociobiology* **47**:873-890.
- Axelrod, R. and W. D. Hamilton. 1981. The Evolution of Cooperation. *Science* **211**:1390-1396.
- Bach, C. E. 1991. Direct and indirect interactions between ants (*Pheidole megacephala*), scales, (*Coccus viridis*), and plants (*Pluchea indica*). *Oecologia* **87**:233-239.
- Bayliss, J. and A. Fielding. 2002. Termitophagous foraging by *Pachycondyla analis* (Formicidae, Ponerinae) in a Tanzanian coastal dry forest. *Sociobiology* **39**:103-122.
- Berger, S., M. Wikelski, L. M. Romero, E. K. V. Kalko, and T. Rödl. 2007. Behavioral and physiological adjustments to new predators in an endemic island species, the Galápagos marine iguana. *Hormones and Behavior* **52**:653-663.

- Bhatkar, A. and W. H. Whitcomb. 1970. Artificial diet for rearing various species of ants. *The Florida Entomologist* **53**:229-232.
- Bluthgen, N., G. Gebauer, and K. Fiedler. 2003. Disentangling a rainforest food web using stable isotopes: dietary diversity in a species-rich ant community. *Oecologia* **137**:426-435.
- Bolton, B. 1995. A Taxonomic and Zoogeographical census of the extant ant taxa (Hymenoptera, Formicidae). *Journal of Natural History* **29**:1037-1056.
- Bourguignon, T., M. Leponce, and Y. Roisin. 2009. Insights into the termite assemblage of a neotropical rainforest from the spatio-temporal distribution of flying alates. *Insect Conservation and Diversity* **2**:153-162.
- Brian, M. V. 1965a. Intergeneric competition. Pages 97-102 *in* M. V. Brian, editor. *Social Insect Populations*. Academic Press, LondonNew York.
- Brian, M. V. 1965b. Interspecific competition. Pages 87-96 *in* M. V. Brian, editor. *Social Insect Populations*. Academic Press, LondonNew York.
- Brian, M. V. 1965c. Intraspecific competition. Pages 78-86 *in* M. V. Brian, editor. *Social Insect Populations*. Academic Press, LondonNew York.
- Brian, M. V. 1965d. Structural Limitations. Pages 66-72 *in* M. V. Brian, editor. *Social Insect Populations*. Academic Press, LondonNew York.
- Buczowski, G. and G. Bennett. 2007. Protein marking reveals predation on termites by the woodland ant, *Aphaenogaster rudis*. *Insectes Sociaux* **54**:219.
- Buczowski, G. and G. Bennett. 2008. Behavioral interactions between *Aphaenogaster rudis* (Hymenoptera : Formicidae) and *Reticulitermes flavipes* (Isoptera : Rhinotermitidae): The importance of physical barriers. *Journal of Insect Behavior* **21**:296-305.
- Case, T. J. and M. E. Gilpin. 1974. Interference competition and niche theory. *Proceedings of the National Academy of Sciences of the United States of America* **71**:3073.
- Constantino, R. 2002. The pest termites of South America: taxonomy, distribution and status. *Journal of Applied Entomology-Zeitschrift Fur Angewandte Entomologie* **126**:355-365.
- Corbara, B. and A. Dejean. 2000. Adaptive behavioral flexibility of the ant *Pachycondyla analis* (=Megaponera foetens) (Formicidae : Ponerinae) during prey capture. *Sociobiology* **36**:465-483.

- Cornelius, M. L. and J. K. Grace. 1995. Laboratory Evaluations of Interactions of 3 Ant Species with the Formosan Subterranean Termite (Isoptera, Rhinotermitidae). *Sociobiology* **26**:291.
- Cornelius, M. L. and J. K. Grace. 1996. Effect of two ant species (Hymenoptera: Formicidae) on the foraging and survival of the Formosan subterranean termite (Isoptera: Rhinotermitidae). *Environmental Entomology* **25**:85-89.
- Cornelius, M. L. and J. K. Grace. 1997. Effect of termite soldiers on the foraging behavior of *Coptotermes formosanus* (Isoptera: Rhinotermitidae) in the presence of predatory ants. *Sociobiology* **29**:247-253.
- Davidson, D. W. 1997. The role of resource imbalances in the evolutionary ecology of tropical arboreal ants. *Biological Journal of the Linnean Society* **61**:153-181.
- Davidson, D. W. 1998. Resource discovery versus resource domination in ants: a functional mechanism for breaking the trade-off. *Ecological Entomology* **23**:484-490.
- Davies, R. G., P. Eggleton, D. T. Jones, F. J. Gathorne-Hardy, and L. M. Hernandez. 2003. Evolution of termite functional diversity: analysis and synthesis of local ecological and regional influences on local species richness. *Journal of Biogeography* **30**:847-877.
- Dejean, A., B. Bolton, and J. L. Durand. 1997. *Cubitermes subarquatus* termitaries as shelters for soil fauna in African rainforests. *Journal of Natural History* **31**:1289-1302.
- Dejean, A., J. L. Durand, and B. Bolton. 1996. Ants inhabiting *Cubitermes* termitaries in African rain forests. *Biotropica* **28**:701-713.
- Dejean, A. and R. Feneron. 1999. Predatory behaviour in the ponerine ant, *Centromyrmex bequaerti*: a case of termitolesty. *Behavioural Processes* **47**:125-133.
- Dejean, A., M. Kenne, and C. S. Moreau. 2007. Predatory abilities favour the success of the invasive ant *Pheidole megacephala* in an introduced area. *Journal of Applied Entomology* **131**:625-629.
- Dejean, A., J. Lachaud, and G. Beugnon. 1993. Efficiency in the exploitation of patchy environments by the Ponerine and *Paltothyreus tarsatus*: An ecological consequence of the flexibility of prey capture behavior. *Journal of Ethology* **11**:43-53.
- Eggleton, P. 2000. Global patterns of termite diversity. Pages 25-51 in T. Abe, D. E. Bignell, and M. Higashi, editors. *Termites: Evolution, Sociality, Symbiosis, Ecology*. Kluwer Academic Publisher, Dordrecht/Boston/London.

- Eggleton, P., D. E. Bignell, W. A. Sands, N. A. Mawdsley, Lawton, T. G. Wood, and N. C. Bignell. 1996. The diversity, abundance and biomass of termites under differing levels of disturbance in the Mbalmayo Forest Reserve, southern Cameroon. *Philosophical Transactions of the Royal Society of London Series B-Biological Sciences* **351**:51-68.
- Fellers, J. H. 1987. Interference and Exploitation in a guild of woodland ants. *Ecology* **68**:1466-1478.
- Garcia Perez, J. A., A. Blanco Pinon, R. Mercado Hernandez, and M. Badii. 1997. Predation of *Pachycondyla harpax* Fabr. (Hymenoptera: Ponerinae) on *Gnathamitermes tubiformans* Buckley (Isoptera: Termitidae) under conditions of captivity. *Southwestern Entomologist* **22**:345-353.
- Gause, G. F. 1934. Experimental Analysis of Vito Volterra's Mathematical Theory of the Struggle for Existence. *Science* **79**:16-17.
- Gause, G. F., O. K. Nastukova, and W. W. Alpatov. 1934. The influence of biologically conditioned media on the growth of a mixed population of *Paramecium caudatum* and *P. aurelia*. *Journal of animal Ecology* **3**:220-228.
- Grover, C. D., A. D. Kay, J. A. Monson, T. C. Marsh, and D. A. Holway. 2007. Linking nutrition and behavioural dominance: carbohydrate scarcity limits aggression and activity in Argentine ants. *Proceedings of the Royal Society B-Biological Sciences* **274**:2951-2957.
- Guénard, B. unpublished data. The Biogeography of *Pachycondyla chinensis*: Ant community structure where *P. chinensis* is introduced. North Carolina State University, Raleigh.
- Guénard, B., R. R. Dunn, M. Fitzpatrick, C. Jenkins, N. McCoy, N. J. Sanders, and M. D. Weiser. 2009. Global Ants. US Department of Energy and a NASA biodiversity grant.
- Hardin, G. 1960. Competitive Exclusion Principle. *Science* **131**:1292-1297.
- Helms, K. R. and S. B. Vinson. 2002. Widespread association of the invasive ant *Solenopsis invicta* with an invasive mealybug. *Ecology* **83**:2425-2438.
- Helms, K. R. and S. B. Vinson. 2008. Plant resources and colony growth in an invasive ant: The importance of honeydew-producing Hemiptera in carbohydrate transfer across trophic levels. *Environmental Entomology* **37**:487-493.

- Herbers, J. M. 1989. Community structure in North temperate ants - Temporal and Spatial variation. *Oecologia* **81**:201-211.
- Hölldobler, B. and J. F. A. Traniello. 1980. The pygidial gland and chemical recruitment communication in *Pachycondyla* (= *Termitopone*) *laevigata*. *Journal of Chemical Ecology* **6**:883-893.
- Hölldobler, B. and E. O. Wilson. 1990. *The Ants*. The Belknap Press of Harvard University Press, Cambridge, Massachusetts.
- Holway, D. and T. Case. 2000. Mechanisms of dispersed central-place foraging in polydomous colonies of the Argentine ant. *Animal behavior* **59**:433-441.
- Holway, D. A. 1999. Competitive mechanisms underlying the displacement of native ants by the invasive argentine ant. *Ecology* **80**:238.
- Holway, D. A., L. Lach, A. V. Suarez, N. D. Tsutsui, and T. J. Case. 2002. The causes and consequences of ant invasions. *Annual Review of Ecology and Systematics* **33**:181-233.
- Human, K. G. and D. M. Gordon. 1996. Exploitation and interference competition between the invasive Argentine ant, *Linepithema humile*, and native ant species. *Oecologia* **105**:405.
- Jaffe, K., C. Ramos, and S. Issa. 1995. Trophic Interactions between Ants and Termites That Share Common Nests. *Annals of the Entomological Society of America* **88**:328.
- Janzen, D. H. 1973. Evolution of polygonous obligate acacia-ants in Western Mexico. *Journal of Animal Ecology* **42**:727-750.
- Jones, T. H., M. S. Blum, and H. M. Fales. 1982. Ant venom alkaloids from *Solenopsis* and *Monomorium* species - Recent developments. *Tetrahedron* **38**:1949-1958.
- Kiesecker, J. M. and A. R. Blaustein. 1997. Population differences in responses of red-legged frogs (*Rana aurora*) to introduced bullfrogs. *Ecology* **78**:1752-1760.
- King, J. R. and W. R. Tschinkel. 2008. Experimental evidence that human impacts drive fire ant invasions and ecological change. *Proceedings of the National Academy of Sciences of the United States of America* **105**:20339-20343.
- Kubota, M., H. T. Imai, M. Kondo, K. Onoyama, K. Ogata, M. Terayama, and M. Yoshimura. 2003. Japanese Ant image Database. Japanese Ant Database Group (JADG).

- Lach, L. 2005. Interference and exploitation competition of three nectar-thieving invasive ant species. *Insectes Sociaux* **52**:257.
- Leal, I. R. and P. S. Oliveira. 1995. Behavioral Ecology of the Neotropical Termite Hunting Ant *Pachycondyla* (= *Termitopone*) *marginata* - Colony Founding, Group-Raiding and Migratory Patterns. *Behavioral Ecology and Sociobiology* **37**:373.
- Lepage, M. G. 1981. Predation of *Megaponera foetens* (F) on Macrotermitinae Foraging Populations in a Semiarid Ecosystem (Kajiado-Kenya). *Insectes Sociaux* **28**:247.
- Lo, N. and T. A. Evans. 2007. Phylogenetic diversity of the intracellular symbiont *Wolbachia* in termites. *Molecular Phylogenetics and Evolution* **44**:461-466.
- Lodge, D. M. 1993. Biological invasions; lessons for ecology. *Trends in Ecology & Evolution* **8**:133-137.
- Long, C. E., B. L. Thorne, and N. L. Breisch. 2003. Termite colony ontogeny: a long-term assessment of reproductive lifespan, caste ratios and colony size in *Reticulitermes flavipes* (Isoptera : Rhinotermitidae). *Bulletin of Entomological Research* **93**:439-445.
- Long, C. E., B. L. Thorne, and N. L. Breisch. 2007. Termite colony ontogeny: supplemental data in the long-term assessment of reproductive lifespan, female neotenic production and colony size in *Reticulitermes flavipes* (Isoptera : Rhinotermitidae). *Bulletin of Entomological Research* **97**:321-325.
- Longhurst, C., R. A. Johnson, and T. G. Wood. 1979. Foraging, Recruitment and Predation by *Decamorium uelense* (Sanstchi) (Formicidae, Myrmicinae) on Termites in Southern Guinea Savanna, Nigeria. *Oecologia* **38**:83.
- Luchetti, A., M. Marini, and B. Mantovani. 2007. Filling the European gap: Biosystematics of the eusocial system *Reticulitermes* (Isoptera, Rhinotermitidae) in the Balkanic Peninsula and Aegean area. *Molecular Phylogenetics and Evolution* **45**:377-383.
- Maloney, R. F. and I. G. Mclean. 1995. Historical and Experimental Learned Predator Recognition in Free-Living New-Zealand Robins. *Animal Behaviour* **50**:1193-1201.
- Marini, M. and B. Mantovani. 2002. Molecular relationships among European samples of *Reticulitermes* (Isoptera, Rhinotermitidae). *Molecular Phylogenetics and Evolution* **22**:454-459.
- Martin, P. and P. Bateson. 1993. *Measuring Behavior an introductory guide*. Second edition. Cambridge University Press.

- Maschwitz, U. and P. Schonegge. 1983. Forage Communication, Nest Moving Recruitment, and Prey Specialization in the Oriental Ponerine *Leptogenys chinensis*. *Oecologia* **57**:175.
- Matsuura, K. 2002. Colony-level stabilization of soldier head width for head-plug defense in the termite *Reticulitermes speratus* (Isoptera : Rhinotermitidae). *Behavioral Ecology and Sociobiology* **51**:172-179.
- Matsuura, K., E. L. Vargo, K. Kawatsu, P. E. Labadie, H. Nakano, T. Yashiro, and K. Tsuji. 2009. Queen Succession Through Asexual Reproduction in Termites. *Science* **323**:1687-1687.
- McGlynn, T. P. 1999. The worldwide transfer of ants: geographical distribution and ecological invasions. *Journal of Biogeography* **26**:535-548.
- Medeiros, L. C. D., A. G. Bandeira, and C. Martius. 1999. Termite swarming in the northeastern Atlantic rain forest of Brazil. *Studies on Neotropical Fauna and Environment* **34**:76-87.
- Mill, A. E. 1984. Predation by the Ponerine Ant *Pachycondyla commutata* on Termites of the Genus *Syntermes* in Amazonian Rain-Forest. *Journal of Natural History* **18**:405.
- Mooney, H. A. and E. E. Cleland. 2001. The evolutionary impact of invasive species. *Proceedings of the National Academy of Sciences of the United States of America* **98**:5446.
- Morales, M. A. 2000. Mechanisms and Density Dependence of Benefit in an Ant-Membracid Mutualism. *Ecology* **81**:482.
- Morrison, L. W. 2000. Mechanisms of interspecific competition among an invasive and two native fire ants. *Oikos* **90**:238-252.
- Nelder, M., E. Paysen, P. Zungoli, and E. Benson. 2006. Emergence of the introduced ant *Pachycondyla chinensis* (Formicidae : Ponerinae) as a public health threat in the southeastern United States. *Journal of Medical Entomology* **43**:1094-1098.
- Neoh, K. B. and C. Y. Lee. 2009. Flight Activity and Flight Phenology of the Asian Subterranean Termite, *Coptotermes gestroi* (Blattodea: Rhinotermitidae). *Sociobiology* **54**:521-530.
- Offenberg, J. 2001. Balancing between mutualism and exploitation: The symbiotic interaction between *Lasius* ants and aphids. *Behavioral Ecology and Sociobiology* **49**:304.

- Overall, W. and A. Bandeira. 1985. Note on the habits of *Cylindromyrmex striatus* Mayr, 1870; Amazonia (Formicidae, Ponerinae). *Revista Brasileira de Entomologia* **29**:521-522.
- Palmer, T. M. 2003. Spatial habitat heterogeneity influences competition and coexistence in an African acacia ant guild. *Ecology* **84**:2843-2855.
- Park, Y. C., O. Kitade, M. Schwarz, J. P. Kim, and W. Kim. 2006. Intraspecific molecular phylogeny, genetic variation and phylogeography of *Reticulitermes speratus* (Isoptera : Rhinotermitidae). *Molecules and Cells* **21**:89-103.
- Paul, J. and F. Roces. 2003. Fluid intake rates in ants correlate with their feeding habits. *Journal of Insect Physiology* **49**:347-357.
- Piek, T., B. Hue, P. Mantel, T. Nakajima, and J. O. Schmidt. 1991. Pharmacological characterization and chemical fractionation of the venom of the ponerine ant, *Paraponera clavata* (F). *Comparative Biochemistry and Physiology C-Pharmacology Toxicology & Endocrinology* **99**:481-486.
- Prestwich, G. D. 1984. Defense mechanisms of termites. *Annual Review of Entomology* **29**:201-232.
- Quinlan, R. J. and J. M. Cherrett. 1979. Role of fungus in the diet of the leaf-cutting ant *Atta cephalotes* (L). *Ecological Entomology* **4**:151-160.
- Rebello, A. M. C. and C. Martius. 1994. Dispersal flights of termites in Amazonian forests (Isoptera). *Sociobiology* **24**:127-146.
- Reitz, S. R. and J. T. Trumble. 2002. Competitive Displacement Among Insects and Arachnids. *Annual Review of Entomology* **47**:435.
- Rojas, M. G. and J. A. Morales-Ramos. 2001. Bait matrix for delivery of chitin synthesis inhibitors to the formosan subterranean termite (Isoptera : Rhinotermitidae). *Journal of Economic Entomology* **94**:506-510.
- Rowles, A. D. and J. Silverman. 2009. Carbohydrate supply limits invasion of natural communities by Argentine ants. *Oecologia* **161**:161-171.
- Sanders, N. J. and D. M. Gordon. 2003. Resource-dependent interactions and the organization of desert ant communities. *Ecology* **84**:1024-1031.
- SAS-v7.0. 2007. Statistical discovery from SAS. *in* SAS, editor. JMP, Cary, NC, USA.
- Scheffrahn, R. H., J. Krecek, B. Maharajh, N. Y. Su, J. A. Chase, J. R. Mangold, A. L. Szalanski, J. W. Austin, and J. Nixon. 2004. Establishment of the African termite, *Coptotermes sjostedti* (Isoptera : Rhinotermitidae), on the Island of Guadeloupe, French West Indies. *Annals of the Entomological Society of America* **97**:872-876.

- Smallwood, J. 1982. The effect of shade and competition on emigration rate in the ant *Aphaenogaster rudis*. *Ecology* **63**:124-134.
- Smith, M. R. 1934. Ponerine ants of the Genus *Euponera* in the United States. *Annals Entomological Society of America* **27**:557-564.
- Sogbesan, A. O. and A. A. A. Ugwumba. 2008. Nutritional values of some non-conventional animal protein feedstuffs used as fishmeal supplement in aquaculture practices in Nigeria. *Turkish Journal of Fisheries and Aquatic Sciences* **8**:159-164.
- Southerland, M. T. 1988. The effects of temperature and food on the growth of laboratory colonies of *Aphaenogaster rudis* Emery (Hymenoptera, Formicidae). *Insectes Sociaux* **35**:304-309.
- Suarez, A. V., D. A. Holway, and P. S. Ward. 2005. The role of opportunity in the unintentional introduction of nonnative ants. *Proceedings of the National Academy of Sciences of the United States of America* **102**:17032-17035.
- Suzzoni, J. P., B. Schatz, and A. Dejean. 2000. Essential and alternative prey in a ponerine ant: variations according to the colony life cycle. *Comptes Rendus De L Academie Des Sciences Serie Iii-Sciences De La Vie-Life Sciences* **323**:1003-1008.
- Szalanski, A. L., J. W. Austin, and C. B. Ovens. 2003. Identification of *Reticulitermes* spp. (Isoptera : Reticulitermatidae) from south central United States by PCR-RFLP. *Journal of Economic Entomology* **96**:1514-1519.
- Szalanski, A. L., R. H. Scheffrahn, J. W. Austin, J. Krecek, and N. Y. Su. 2004. Molecular Phylogeny and biogeography of *Heterotermes* (Isoptera : Rhinotermitidae) in the West Indies. *Annals of the Entomological Society of America* **97**:556-566.
- Tanner, C. J. and F. R. Adler. 2009. To fight or not to fight: context-dependent interspecific aggression in competing ants. *Animal Behaviour* **77**:297-305.
- Tillberg, C. V., D. A. Holway, E. G. LeBrun, and A. V. Suarez. 2007. Trophic ecology of invasive Argentine ants in their native and introduced ranges. *Proceedings of the National Academy of Sciences of the United States of America* **104**:20856-20861.
- Tschinkel, W. R. 2003. Subterranean ant nests: trace fossils past and future? *Palaeogeography Palaeoclimatology Palaeoecology* **192**:321-333.
- Walsh, P. S., D. A. Metzger, and R. Higuchi. 1991. Chelex-100 as a medium for simple extraction of DNA for PCR-based typing from forensic material. *Biotechniques* **10**:506-513.

- Watt, A. D., N. E. Stork, P. Eggleton, D. Srivastava, B. Bolton, T. B. Larsen, M. J. Brendell, and D. E. Bigneli. 1997. Impact of forest loss and regeneration on insect abundance and diversity. Pages 273-286 in A. D. Watt and M. D. Hunter, editors. *Forests and insects*. Chapman and Hall, London.
- Way, M. J. 1963. Mutualism between ants and honeydew-producing Homoptera. *Annual Review of Entomology* **8**:307-329.
- Weesner, F. M. 1965. *The Termites of the United States: A handbook*. The National Pest Control Association, Elizabeth, New Jersey.
- Wheeler, W. M. 1930. A list of the known Chinese Ants. *Peking Natural History Bulletin* **5**:53-81.
- Wheeler, W. M. 1936. Ecological relations of ponerine and other ants to termites. *Proceedings on the American Academy of Arts and Sciences* **71**:159-243.
- Wildy, E. L. and A. R. Blaustein. 2001. Learned recognition of intraspecific predators in larval long-toed salamanders *Ambystoma macrodactylum*. *Ethology* **107**:479-493.
- Wilson, E. O. and W. L. J. Brown. 1984. Behavior of the cryptobiotic predaceous ant *Eurhopalothrix heliscata*, N. SP. (Hymenoptera: Formicidae: Basicerotini). *Insectes Sociaux* **31**:408-428.
- Wood, T. G. and W. A. Sands. 1978. The Role of Termites in Ecosystems. Pages 245-292 in M. V. Brian, editor. *Production ecology of Ants and Termites*. Cambridge University Press, Cambridge.
- Ye, W. M., C. Y. Lee, R. H. Scheffrahn, J. M. Aleong, N. Y. Su, G. W. Bennett, and M. E. Scharf. 2004. Phylogenetic relationships of nearctic *Reticulitermes* species (Isoptera : Rhinotermitidae) with particular reference to *Reticulitermes arenicola* Goellner. *Molecular Phylogenetics and Evolution* **30**:815-822.
- Yeap, B. K., A. S. Othman, and C. Y. Lee. 2009. Molecular Systematics of *Coptotermes* (Isoptera: Rhinotermitidae) From East Asia and Australia. *Annals of the Entomological Society of America* **102**:1077-1090.
- Zee, J. and D. A. Holway. 2006. Nest raiding by the invasive Argentine ant on colonies of the harvester ant, *Pogonomyrmex subnitidus*. *Insectes Sociaux* **53**:161-167.
- Zungoli, P., E. Paysen, E. Benson, and J. Nauman. 2005. Colony and habitat characteristics of *Pachycondyla chinensis* (Hymenoptera: Formicidae).in *Proceedings of the fifth*

International Conference on Urban Pests. Perniagaan Ph'ng @ P&Y Design Network,  
Malaysia.

## APPENDIX

**Table 1 Response of *P. chinensis* to different potential prey. Means (X) are displayed for data analyzed using an ANOVA, while percentages are displayed for Chi2 data. Levels per column not connected by the same letter are significantly different (Tukey-Kramer HSD).**

Prey Item (n=25)	D <sub>t</sub> (X(sec.))	E (X)	A (%Stung)	K (%killed)	K <sub>t</sub> (X(sec.))	R (%retrieved)	R <sub>t</sub> (X(sec.))	F (X)	G (X)	P <sub>a</sub> (%Chosen)	K <sub>e</sub> (X)	R <sub>e</sub> (X)
<i>R. virginicus</i> (workers)	24.4 <sup>C</sup>	1.1 <sup>C</sup>	100%	100%	9.1 <sup>B</sup>	100%	49.4 <sup>A</sup>	5.1 <sup>ABC</sup>	1.2 <sup>B</sup>	100%	37.1 <sup>A</sup>	16.6 <sup>A</sup>
Collembola	195 <sup>A</sup>	6 <sup>BC</sup>	20%	28%	45.4 <sup>B</sup>	28%	17.9 <sup>A</sup>	5.7 <sup>AB</sup>	1 <sup>B</sup>	20%	34 <sup>AB</sup>	24.7 <sup>A</sup>
Parcoblatta spp.	70.1 <sup>BC</sup>	23.8 <sup>A</sup>	76%	28%	209.8 <sup>A</sup>	8%	60 <sup>A</sup>	6.7 <sup>A</sup>	6.3 <sup>A</sup>	8%	6.5 <sup>BC</sup>	7.88 <sup>AB</sup>
Chilopoda	192.2 <sup>AB</sup>	10.4 <sup>B</sup>	56%	24%	267.8 <sup>A</sup>	16%	65.8 <sup>A</sup>	3.2 <sup>C</sup>	4.5 <sup>A</sup>	16%	1.6 <sup>C</sup>	-2.77 <sup>B</sup>
Elateridae (larvae)	437.1 <sup>AB</sup>	2.4 <sup>C</sup>	8%	0%	N/A	0%	N/A	4 <sup>BC</sup>	N/A	0%	N/A	N/A
Trogossitidae (adults)	339.7 <sup>AB</sup>	2.6 <sup>C</sup>	4%	0%	N/A	0%	N/A	2.9 <sup>C</sup>	N/A	0%	N/A	N/A

**Table 2 Average values for observations of one-on-one interactions between ants and termites. \*For Tukey-Kramer HSD levels not connected by the same letter are significantly different at alpha=0.05.**

Resident	Intruder	Mean Number of Encounters	Percentage of interactions initiated by ant	Mean time to kill since 1 <sup>st</sup> interaction (seconds)	Percentage of time ant aggression = 4 (sting)	Percentage of time Termite killed
		Tukey-Kramer HSD* q=2.6, df=3	Pearson X <sup>2</sup> =129.2, df=9, p<0.0001	Tukey-Kramer HSD* q=2.6, df=3	Pearson X <sup>2</sup> =81.6, df=9, p<0.0001	Pearson X <sup>2</sup> =84.25, df=9, p<0.0001
Termite Worker	Aphaenogaster	3.2 <sup>AB</sup>	13%	120 <sup>A</sup>	6.7%	6.7%
	Pachycondyla	1.3 <sup>B</sup>	96%	14.3 <sup>B</sup>	100%	100%
Termite Soldier	Aphaenogaster	3.9 <sup>AB</sup>	30%	81.8 <sup>A</sup>	27%	16.7%
	Pachycondyla	4.5 <sup>A</sup>	73%	29.5 <sup>B</sup>	87%	83.3%

**Table 3 Values of interactions between heterospecific ants.**

Resident	Intruder	Mean Number of Encounters	Percentage of interactions initiated by resident ant	Mean time to kill since 1 <sup>st</sup> interaction (seconds)	Percentage of time ant aggression = 4 (sting)	Percentage of time Aphaenogaster killed
		t Test=-3.1, df=33, p=0.0019	Pearson X <sup>2</sup> =2.8, df=2, p=0.24	t test=-1.3, df=14, p=0.093	Pearson X <sup>2</sup> =2.44, df=2, p=0.29	Pearson X <sup>2</sup> =2.92, df=2, p=0.23
Pachycondyla	Aphaenogaster	4.8	41%	56.2	70.6%	53%
Aphaenogaster	Pachycondyla	13.5	50%	102	66.7%	28%

Figures:

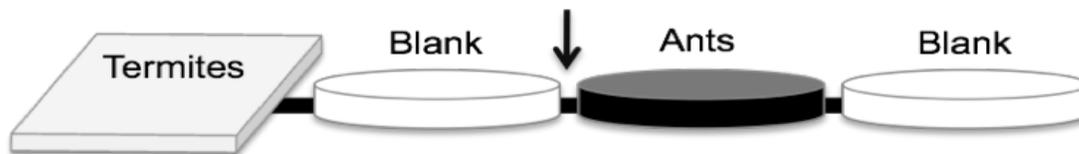


Figure 1 Experimental set up for testing effects of location on predation rate in the lab. The arrow indicates where ant access to termites was blocked during the 24-hour acclimation period.

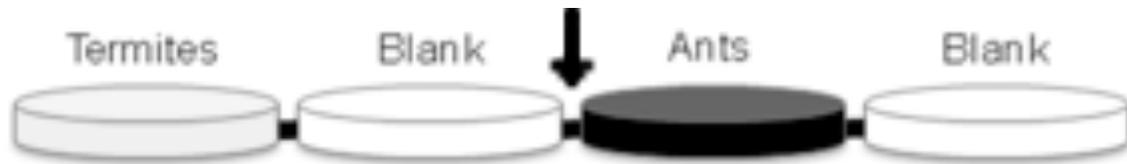


Figure 2 Experimental set up for the observation of each ants ability to prey on termites at different soldier ratios. The arrow indicates the site where ant access to the termites was blocked during the 24-hour acclimation period.

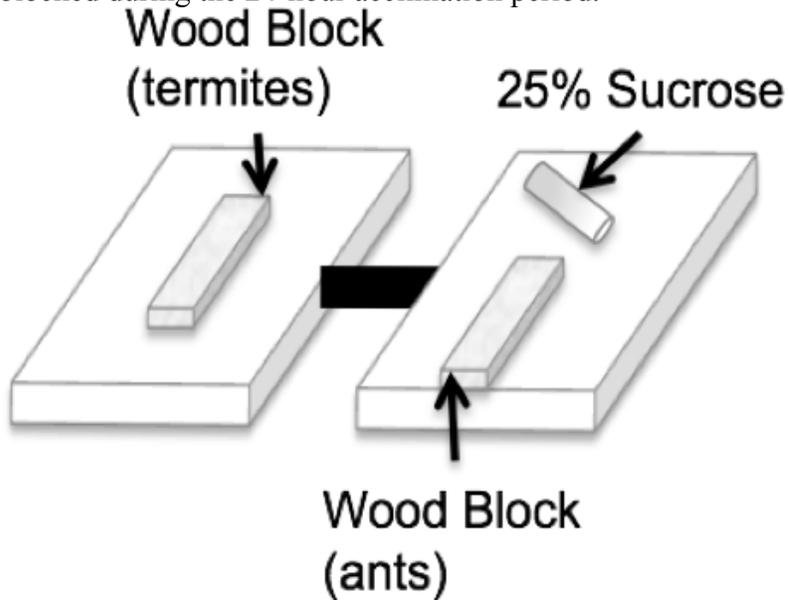


Figure 3 Experimental set up for testing the effectiveness of a natural wood barrier against ant predation. Controls involved petri dishes filled with sand and filter paper instead of wood blocks.

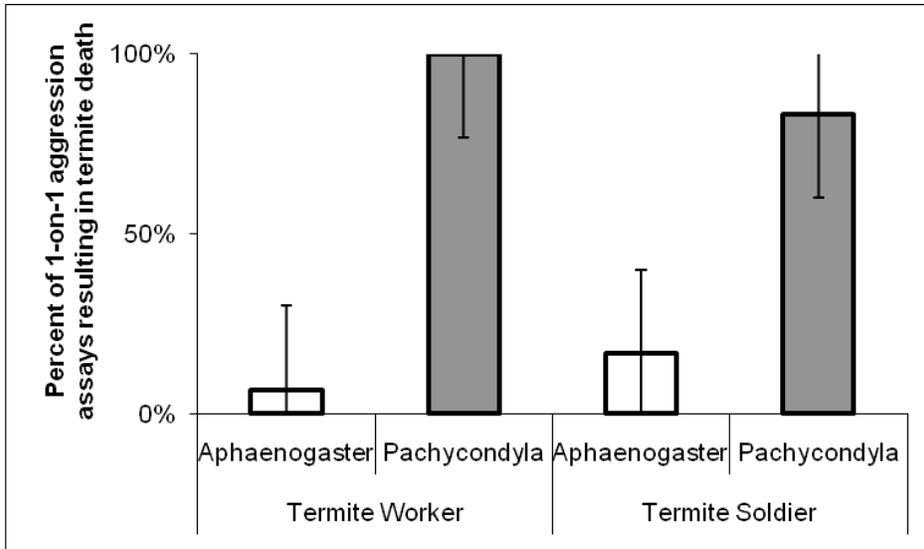


Figure 4 Termite worker (solid columns) and soldier (striped columns) mortality in 1-on-1 aggression assays with *P. chinensis* (shaded columns) and *A. rudis* (open columns).

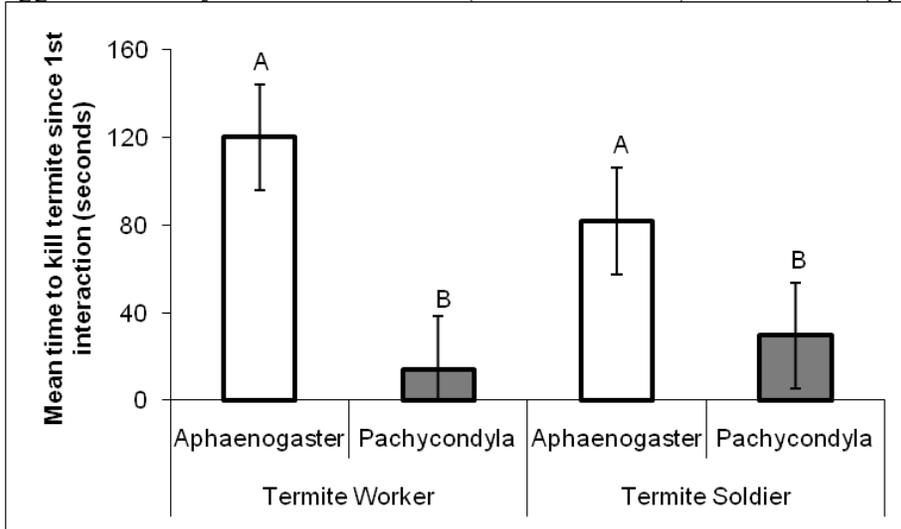


Figure 5 Average time since the first interaction with *A. rudis* (open columns) or *P. chinensis* (shaded columns) for termite worker (solid columns) and soldier (striped columns) death.

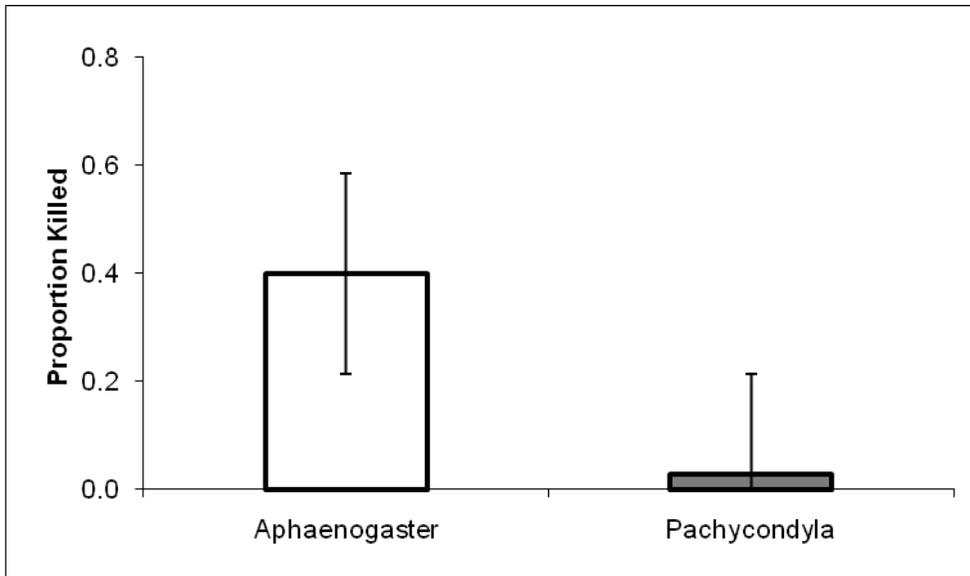


Figure 6 Proportion of 1-on-1 aggression assays resulting in *A. rudis* (open column) and *P. chinensis* (shaded column) death.

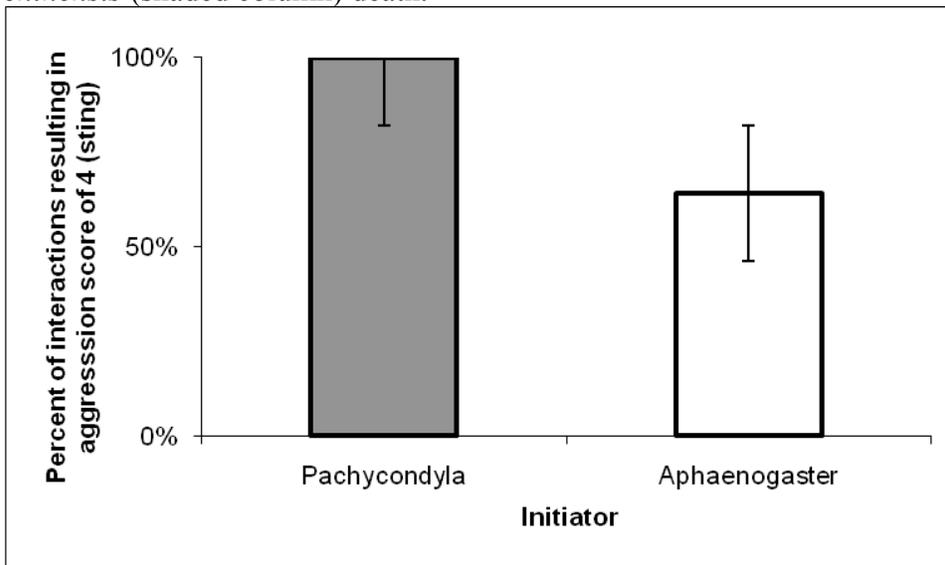


Figure 7 Percentage of interactions between *A. rudis* (open column) and *P. chinensis* (shaded column) resulting in a maximum aggression score of 4 (sting) based upon which ant initiated the interaction.

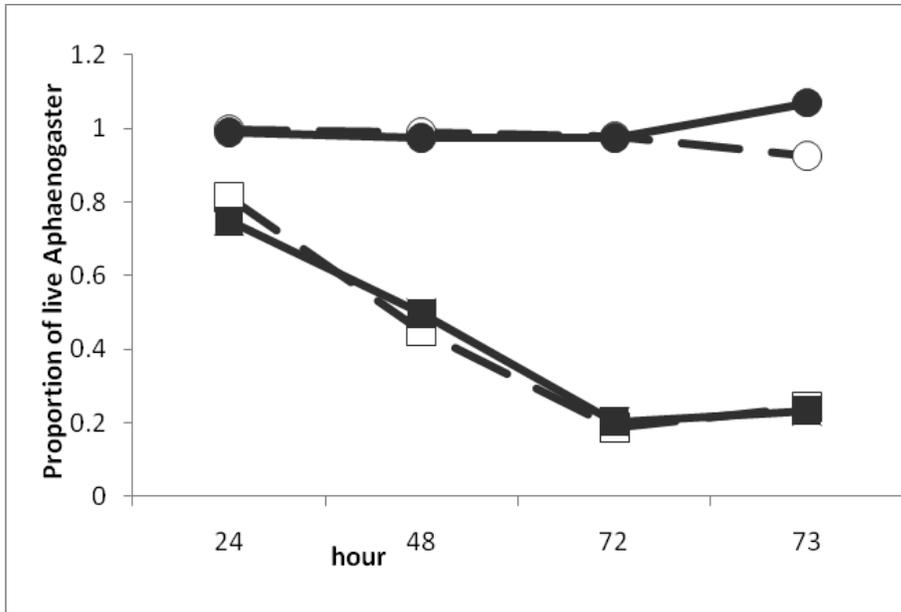
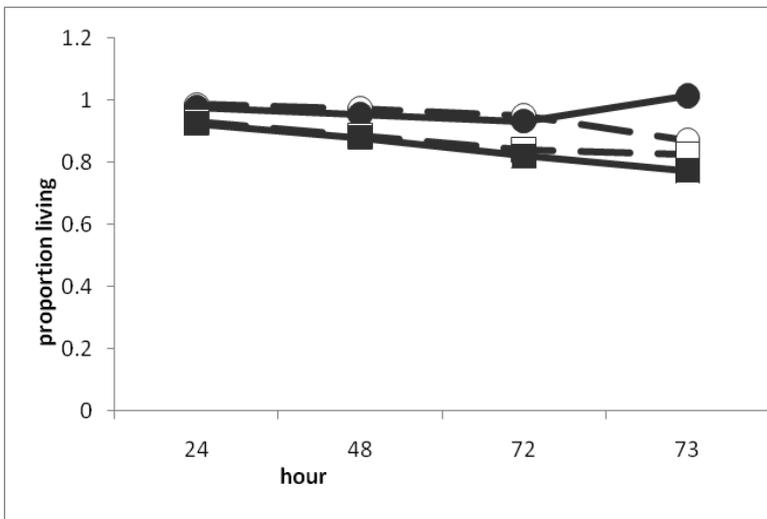


Figure 8 Mortality of (above) *A. rudis* and (below) *P. chinensis* in the presence of termites (shaded points) and the absence (open points) in competition with the heterospecific ant (squares) and without (circles).



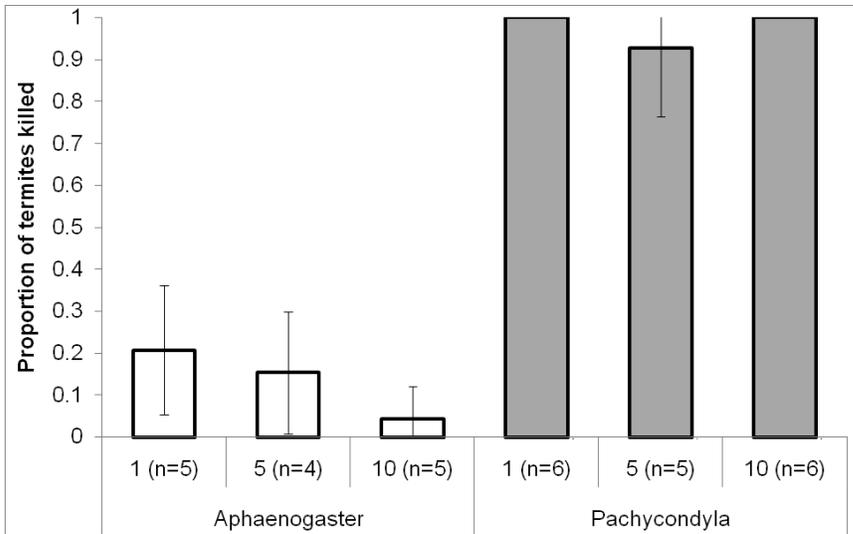


Figure 9 Average proportion of *R. virginicus* mortality at 1, 5, and 10 percent soldier ratio caused by *A. rudis* (open columns) and *P. chinensis* (shaded columns). Control mortality is not shown because values displayed were calculated using the Henderson-Tilton formula. Columns not connected by the same letter are significantly different.

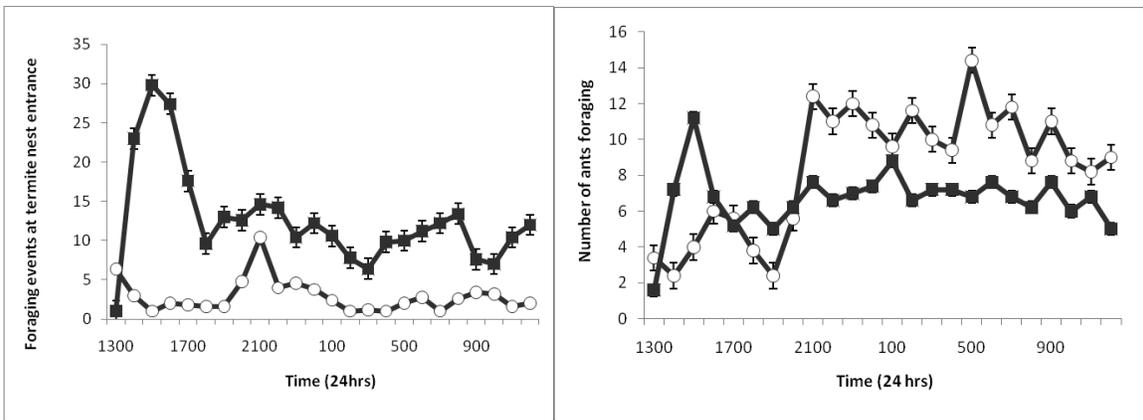


Figure 10 The number of foraging events (above left), foraging ants (above right), and foraging events per ant (below) for *A. rudis* (open circles) and *P. chinensis* (closed squares) in the presence of termites over a 24 hour period (x-axis).

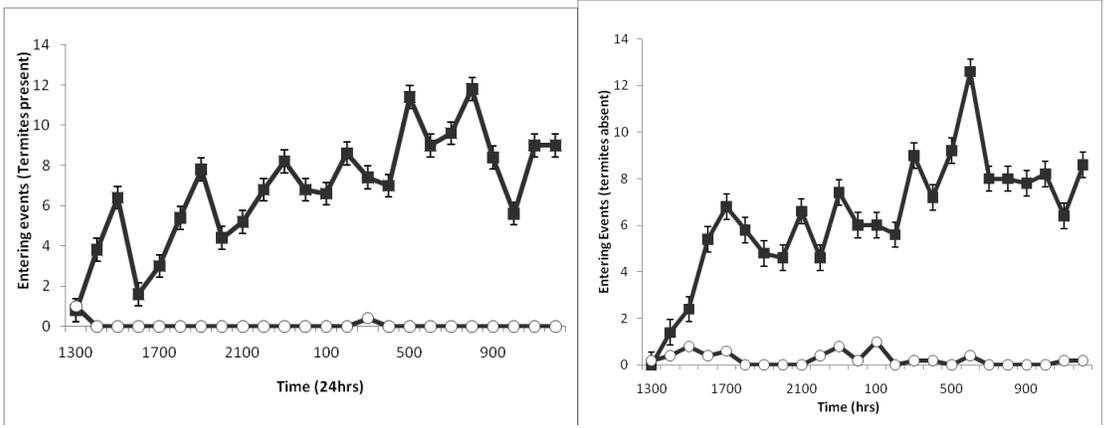


Figure 11 The number of entering events for *P. chinensis* (closed squares) and *A. rudis* (open circles) in the presence (left) and absence (right) of termites.

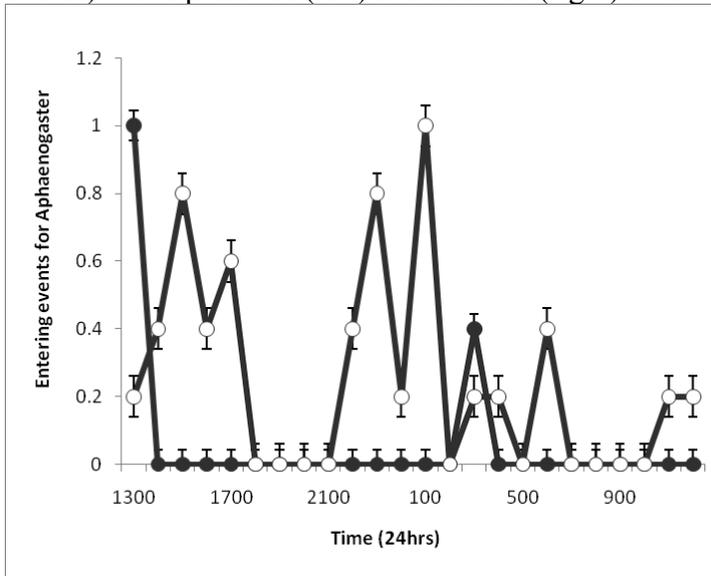


Figure 12. The number of entering events for *A. rudis* in the absence (open circles) and presence (closed circles) of termites over a 24 hour period (x-axis).