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# NO ENEMIES NEEDED: COTTON APHIDS (HEMIPTERA: APHIDIDAE) DIRECTLY BENEFIT FROM RED IMPORTED FIRE ANT (HYMENOPTERA: FORMICIDAE) TENDING

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## ABSTRACT

Honeydew is a carbohydrate-rich solution excreted by phloem-feeding insects such as aphids. Ants often consume this substance and, in return, protect aphids from natural enemies. This indirect benefit of ant-aphid mutualisms to aphids (reduced predation) has been examined extensively. Few studies, however, have quantified the direct benefits that aphids may gain from the mutualism. We conducted greenhouse experiments to estimate the direct benefits that cotton aphids, *Aphis gossypii* (Glover) (Hemiptera: Aphididae), receive from their mutualistic relationship with red imported fire ants, *Solenopsis invicta* (Buren) (Hymenoptera: Formicidae). We compared population growth and alate production between ant-tended and untended aphid colonies in the absence of natural enemies. We found strong evidence that cotton aphids receive direct benefits from their relationship with fire ants. After 12 days, aphid colonies with tending ants were 46% larger than their non-tended counterparts. Alate production, however, was not affected by ant tending, suggesting that a reduction in dispersal did not explain the ant effect on aphid population growth. We hypothesize that the increase in aphid population size results from altered feeding behaviors in the presence of ants. This study suggests that there may be constant selection for this mutualism even in the absence of aphid natural enemies because aphids gain direct benefits.

Key Words: *Solenopsis invicta*, *Aphis gossypii*, ant-aphid mutualism, direct effects

## RESUMEN

El mielcilla es una solución rica en carbohidratos excretada por los insectos que se alimentan del floema como los áfidos. Las hormigas suelen consumir esta sustancia y a cambio, protegen a los áfidos de los enemigos naturales. Este beneficio indirecto para los áfidos del mutualismo de las hormigas y áfidos (reducción de la depredación) ha sido examinada exhaustivamente. Pocos estudios, sin embargo, han cuantificado los beneficios directos que los áfidos pueden obtener del mutualismo. Hemos realizados experimentos de invernadero para estimar los beneficios directos que recibe el áfido de algodón, *Aphis gossypii* (Glover) de su relación mutualística con la hormiga de fuego roja importada, *Solenopsis invicta* (Buren). Se comparó el crecimiento de la población y la producción de alados (áfidos con alas) entre las colonias de áfidos cuidados y no cuidados por hormigas en la ausencia de enemigos naturales. Hemos encontrado fuerte evidencia de que los áfidos de algodón reciben beneficios directos de su relación con las hormigas de fuego. Después de 12 días, las colonias de áfidos cuidados por las hormigas fueron 46% más grandes que sus contrapartes no-cuidados. La producción de áfidos alados, sin embargo, no fue afectada por el cuidado de las hormigas, lo que sugiere que una reducción en la dispersión no explicó el efecto hormiga en crecimiento de la población de áfidos. Se postula que el aumento en el tamaño de la población de áfidos resulta del comportamiento de alimentación alterado en la presencia de hormigas. Este estudio sugiere que puede haber selección constante para este mutualismo aún estando en ausencia de los enemigos naturales de los áfidos porque los áfidos obtienen beneficios directos.

Palabras Clave: *Solenopsis invicta*, *Aphis gossypii*, mutualismo hormiga-áfido, efectos directos

Numerous studies have examined mutualistic interactions between ants and aphids. Ants consume honeydew, a carbohydrate-rich solution excreted by aphids, and in return protect aphids from natural enemies (Way 1963). Few studies, however, have quantified the potential direct benefits aphids receive from tending ants. Direct benefits would provide continuous selection for honeydew-producing insects to form relationships with tending ants and may explain why these relationships are ubiquitous in nature.

Tending ants may directly affect aphid population growth through several mechanisms. Ants can reduce aphid dispersal by reducing the number of alate (winged) individuals within a population. Previous studies intriguingly suggest that ants chemically inhibit aphid wing formation (Kleinjan & Mittler 1975) whereas others suggest ants physically remove wings from alate individuals (Kunkel 1973). A reduction in alates could increase aphid populations by reducing dispersal. The presence of ants may also affect aphid longevity and fecundity. For instance, Flatt & Weisser (2000) observed that when aphid predators were excluded, individual aphids (*Metopeurum fuscoviride* Stroyan) lived significantly longer and matured faster when tended by the black garden ant, *Lasius niger* L. Furthermore, tended aphids produced on average 63 more offspring than unattended aphids. In addition, ant tending may allow aphids to feed at optimal rates due to reduced aphid predator abundance. Increased feeding efficiency can increase resources allocated towards reproduction thereby increasing population growth (Slansky & Scriber 1985). However, Stadler & Dixon (1998) observed black bean aphids, *Aphis fabae* (Scopoli), experience adverse effects when tended by *Lasius niger*. Aphids experienced reductions in growth rates and gonad size when tended by ants suggesting increased feeding rates reduce the efficiency of nutrient assimilation.

Lastly, aphids may receive hygienic benefits from honeydew removal by ants. Excessive honeydew accumulation can result in pathogens such as sooty mold. Sooty mold, a fungus that grows on honeydew, creates a black layer on the upper side of leaves, thereby decreasing photosynthesis and causing leaf abscission (Wood et al. 1988). Sooty mold growth can negatively impact herbivores, including aphids, by reducing host plant quality (Way 1963, Wood et al. 1988). Therefore honeydew removal by ants may benefit aphids by reducing fungal infections and maintaining host plant quality.

Cotton aphids (*Aphis gossypii*) (Glover) are pests of several agricultural crops including cotton (*Gossypium hirsutum* L.), cucurbits, okra (*Abelmoschus esculentus* L.), and tomato (*Lycopersicon esculentum*) (Miller) (Ebert & Cartwright 1997). Aphid infestations in cotton reduce boll

weight (Fuchs & Minzenmayer 1995) and honeydew accumulation can result in additional economic damage by contaminating and reducing the quality of cotton lint (Carter 1992). The red imported fire ant (*Solenopsis invicta*) (Buren) was unintentionally introduced to North America approximately 80 years ago and has spread to at least 15 states. Several studies have quantified the indirect benefits that aphids receive as a result of fire ant protection from natural enemies (e.g., Kaplan & Eubanks 2002, 2005). This study used greenhouse experiments to examine the direct benefits that cotton aphids potentially receive from their mutualistic relationship with fire ants in the absence of natural enemies.

## MATERIALS AND METHODS

To examine the direct effects of ants on aphid population growth and alate production greenhouse experiments were conducted on the campus of Auburn University, Auburn, Alabama, during July 2005. Fifty-six cages (75 × 30 × 30 cm) were constructed from PVC pipe framing and mesh mosquito netting and placed around a 37 liter plastic container filled with potting soil. A cotton plant with 5-6 true leaves (~1 m tall) was placed in each cage. One-hundred apterous cotton aphids of similar coloration and size were transferred from a greenhouse colony to each caged plant and allowed to acclimate for 24 hrs. Field collected fire ant colonies (~500 ants) were added to half (28) of the cages. The inside and outside rim of plastic containers were lined with liquid polytetrafluoroethylene (Teflon®) to exclude ants from entering ant absent treatments and escaping from ant treatments. Ants immediately began tending aphids and consuming honeydew. Starting 5 days after experimental setup and every other day thereafter the upper and lower surface of each leaf, branch, and stem was surveyed and the total number of apterous and alate aphids on each plant recorded. Repeated measures ANOVA (Proc GLM; all statistical analyses were performed using SAS, Version 9.1 [SAS Institute 2002]) was conducted to compare aphid colony growth and alate production.

## RESULTS

We found strong evidence that cotton aphids receive additional direct benefits from their mutualistic relationship with fire ants in the absence of natural enemies. After 7 days, aphid colonies tended by ants were 35% larger than unattended colonies ( $F_{3,158} = 2.02, P = 0.0455$ ). This trend continued throughout the remainder of the experiment and by day 12, aphid colonies with ants were 46% larger than aphid colonies without ants ( $F_{3,158} = 7.19, P < 0.0001$ ) (Fig. 1). No apterous

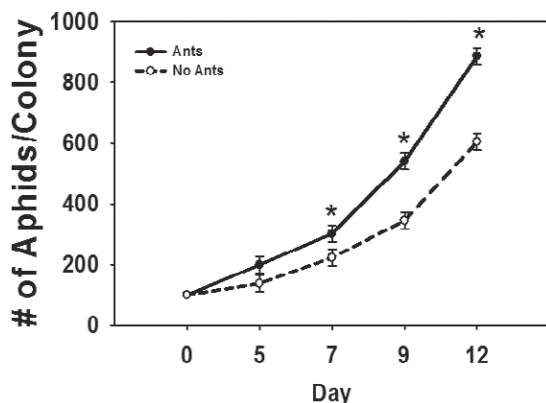


Fig. 1. The direct effects of fire ants on cotton aphid population size (means  $\pm$  SE) in the absence of natural enemies. An asterisk indicates means were significantly different on sampling dates at the  $P = 0.05$  level.

aphid dispersal was observed throughout the experiment. Alate abundance remained low in both treatments throughout the entire experiment and no significance difference was observed between ant tended and untended aphids on any sampling date. At the conclusion of the study ant tended aphids averaged 8.87 alates per colony compared with 9.03 alates in untended colonies ( $F_{3, 158} = 0.08$ ,  $P = 0.9402$ ).

#### DISCUSSION

Tending ants may alter aphid behavior resulting in increased colony growth. For instance, aphid phloem consumption rate depends on duration and frequency of feeding probes (Risebrow & Dixon 1987) and aphids have the ability to alter their feeding rates (Dixon 1963). Additionally, ant tending may signal a reduction in predation risk to aphids, resulting in decreased antipredatory behaviors. Tending ants may encourage aphids to feed continuously, thereby increasing feeding efficiency (Rauch et al. 2002) leading to increased resource uptake and higher reproduction rates. Although few studies have examined this, some evidence does suggest that ant tending affects aphid feeding rates. Banks & Nixon (1958) observed increased honeydew production from tended ants and concluded that ants stimulate aphid feeding. Likewise, Brenton & Addicott (1992) suggest, *Aphis varians* (Patch), increases its feeding rate when tended by the ant *Formica cinera* (Wheeler). Further, aphids often respond to predators or alarm pheromones by dropping from host plants (Losey & Denno 1998). Dropping from host plants can increase individual aphid survival but can decrease population growth due to lost foraging opportunities (Fill et al. 2012). Costly antipredator behaviors; however, can not explain the results of our study because aphid

natural enemies were completely excluded from the aphid colonies.

In addition to reduced antipredatory behavior, honeydew removal by ants likely provides an important hygienic service to aphids. Honeydew accumulation can increase the incidence of pathogens, such as sooty mold, and decrease host plant quality by reducing available photosynthetic leaf area. Bach (1991) observed reduced honeydew and sooty mold accumulation on plants with ants compared with plants without ants. When ants were excluded, 89.5% of leaves contained honeydew; whereas only 6% contained honeydew when ants were present. Additionally, the presence of ants reduced sooty mold growth by 20% in this study. In the absence of ants, Fokkema et al. (1983) observed a tenfold increase in fungal pathogens, such as sooty mold, due to excessive honeydew build up. However due to the short experimental duration and low aphid densities, we observed no evidence of sooty mold contamination in either treatment.

The presence of ants had no effect on aphid alate production. In fact, alate production was very low in both treatments. Previous studies suggest a reduction in alates when ants tend aphids (Kleinjan & Mittler 1975; Tilles & Wood 1982). However, alate production is also inversely correlated with host plant quality (Dixon 1998). Plants in our experiment were healthy and lacked other herbivores. Therefore aphids may not have produced alates regardless of the presence or absence of ants due to the high quality of host plants used in this experiment.

This study is one of the first to document direct benefits to aphids from ant tending. In addition to the indirect benefits provided by ants (protection from natural enemies), aphids receive direct benefits such as increased population growth when fire ants were present. These additional direct benefits likely increase selection pressure for this mutualistic relationship. Future studies quantifying costs and benefits of mutualistic interactions should evaluate direct as well as indirect benefits. Additional studies should also attempt to elucidate the exact mechanisms underlying the direct benefits aphids receive from tending ants.

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