

May 2008

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Use of shade trees to reduce leaf herbivory of *Coffea arabica* by *Atta cephalotes* (Formicidae) on farms in Cañitas, Puntarenas, Costa Rica

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ABSTRACT

The addition of a specific group of rapid-growth shade tree species to coffee in monoculture, or the inclusion of these species in shade-grown coffee, may reduce leaf herbivory by *Atta cephalotes*. This study was conducted in Cañitas, Costa Rica on three plots—shade-grown, monoculture, and no coffee—within coffee plantations, each of which had a separate *A. cephalotes* colony. After observing the selection process of the leaf-cutter ants among four non-coffee, shade tree species and two varieties of coffee, Caturra and Katway, results indicated that the ants had no preference(s) of the tested species within any of the tested plots (Shade: K-W = 0.976, $p = 0.98$; Monoculture: K-W = 1.319, $p = 0.971$; No Coffee: K-W = 0.803, $p = 0.992$; All plots: $df = 6$, $n = 35$). The ants more frequently selected *Erythrina lanceolata* at the Shade-Grown Plot than the other two plots (K-W = 7.22, $n = 15$, $df = 2$, $p = 0.027$). These findings may have important implications for both coffee farming and the restoration of biodiversity in monoculture crops.

RESUMEN

La adición de un grupo específico de especies de árboles que dan sombra y crecen rápido al café en monocultivo, o la inclusión de estas especies en café que crece en la sombra, podría reducir la herbivoría de las hojas por *Atta cephalotes*. Este estudio fue conducido en Cañitas, Costa Rica en tres parcelas—crece en la sombra, monocultivo, y no café—dentro de plantaciones de café, cada una de las cuales tenía una colonia diferente de *A. cephalotes*. Después de observar el método de selección por las hormigas entre cuatro especies de árboles que dan sombra que no tenían café y dos variedades de café, Caturra y Katway, los resultados indicaron que las hormigas no tenían preferencias por las especies probados dentro de cada parcela (Sombra: K-W = 0.976, $p = 0.98$; Monocultivo: K-W = 1.319, $p = 0.971$; No café: K-W = 0.803, $p = 0.992$; Todos: $df = 6$, $n = 35$). Las hormigas escogieron más frecuentemente *Erythrina lanceolata* en la parcela de café que crece en la sombra que las otras (K-W = 7.22, $n = 15$, $df = 2$, $p = 0.027$). Estos resultados podrían tener implicaciones importantes para ambos la labranza de café y la restauración de biodiversidad en las cosechas de monoculture.

INTRODUCTION

Coffee is one of the most important and widespread crops in Central America. In Costa Rica, annual exports of coffee exceed \$197 million. Coffee is grown in two ways: “shade-grown” in the understory, surrounded by larger trees that provide the soil with nutrients, and also in monoculture. The latter method removes all trees and replaces old varieties of coffee with new sun-loving varieties (Perfecto and Snelling 1995). The

majority of coffee in Central America is grown in monoculture (Perfecto and Snelling 1995), which reduces biodiversity and increases crop vulnerability to pests. Expensive fossil fuels are used as fertilizers or pesticides, but are difficult to obtain in the tropics (Ewel 1986) and are not environmentally friendly. For this reason, farmers that use shade grown methods benefit from the ability of some non-coffee, shade tree species to return nitrogen to the soil.

Atta cephalotes, a leaf-cutting ant, can be found in many areas in the neotropics. It generally lives below 2000 meters, and can be found in all types of forest (Stevens 1983); they especially like gap areas (Blanton and Ewel 1985). Though originally considered a generalist forager, studies support that *A. cephalotes* attacks plants selectively (Rockwood 1976, Howard 1988). Among the selections of *A. cephalotes* are several Neotropical crops grown in monoculture (Blanton and Ewel 1985), including cassava and coffee. Herbivory by *A. cephalotes* greatly negatively affects net productivity (Rockwood 1972), undoubtedly creating a great concern for farmers.

In addition to crop success, maintaining the original biodiversity of these disturbed areas is extremely important. Changes in biodiversity in agricultural areas and other areas of high human impact have not been studied as extensively as those in more natural habitats. A study of coffee farming in Mexico found traditional coffee growing methods to only partially disturb original forests (Moguel and Toledo 1999). Therefore, these traditional, shade-grown coffee plantations can have high species richness and may be important homes for many organisms, including plants and arthropods. It is, therefore, highly beneficial for both the farmers and the environment to design agricultural growth to mimic natural surrounding growth of the area.

The objective of this study is to examine whether *A. cephalotes* shows preferences among two varieties of *C. arabica* and several shade tree species that could potentially be grown with coffee. A recent study at a farm in San Luis, Costa Rica, studied leaf preferences of *A. cephalotes* with several shade tree species and coffee varieties (Hannemann 2007). Results from this study suggested a preference for the tree species over the coffee. This study only focused on two sites, in one farm: in monoculture coffee and in secondary forest. Based on this previous study it is expected that *A. cephalotes* will show a preference towards non-coffee shade trees, therefore showing that shade trees may offer practical uses for farmers in reducing leaf herbivory of coffee. It may also provide an opportunity to restore some biodiversity to habitats struggling from monoculture practices. I hypothesize that because coffee can support the growth of various basidiomycete fungi (Avelino et al. 2007), the ants will preferentially select the coffee varieties over the shade tree species within each study site.

METHODS

This study was performed on three plots in coffee farms in Cañitas, Costa Rica, from April 15 to May 3, 2008. Each plot investigated a separate ant colony. The Shade-Grown Plot had both varieties of shade-grown *C. arabica*, Caturra and Katway, within close proximity to the ant nest. The Monoculture Plot was approximately three meters from both a plot of monoculture Katway coffee and a plot of monoculture Caturra, and the No-Coffee Plot lacked coffee and was made up of tall grasses and shrubs.

The following shade tree species (Table 1) were selected for this experiment because of their rapid growth and because they are native to the Monteverde area. Extracts were made from all four shade tree species, from Caturra and Katway varieties of *C. arabica*, and also for a control extract, which did not have any plant material.

TABLE 1. Family, Genus and species of the six plants tested.

Species	Family
<i>Erythrina lanceolata</i>	Papilionaceae
<i>Sapium glandulosum</i>	Euphorbiaceae
<i>Citharexylum costaricensis</i>	Verbenaceae
<i>Acnistus arborescens</i>	Solanaceae
<i>C. arabica</i> (Caturra)	Rubiaceae
<i>C. arabica</i> (Katway)	Rubiaceae

Plant extract and oat preparation

Oats are commonly used as a medium for extracts when testing plant preference in leaf-cutter ants. From each plant, five grams of leaves were removed and added to 50 ml of 0.8 M methanol and 1% hydrochloric acid in a 1:1 ratio. Each solution was ground for ten minutes. Each solution was then strained into separate film canisters and each shaken to complete mixing. The control extract was methanol and hydrochloric acid with no leaf matter. Approximately 50 oats were placed in the strainer for each extract application and an eyedropper was used to apply the plant extracts to the oats. The oats were then removed from the strainer and placed on a paper to dry. This was repeated several times to prepare for each day of experimentation. The oats were dried for at least an hour before use.

Extract Preference Tests

The study took place approximately five meters from the nest at each of the three sites. At each plot, five oats from each of the six plant extracts and the control extract were placed separately on to a piece of paper in a line across the ant trail. The quantities removed from the stock of each extract were observed and recorded for two hours or until the supply of one extract ran out. Each time an ant removed an oat from the starting position and carried it toward the colony, the oat was replaced with another oat of the same extract. The plant extracts and control were placed in different positions every day to reduce any bias toward certain positions. This study was conducted five days at each plot, beginning at 8:30 AM. Because the ants frequently depleted the daily supply of one or more extract, daily removal quantities were modified to represent the quantity of oats removed in 60 minutes at a study site.

Statistical Tests

I used Kruskal-Wallis tests to compare the preferences of each extract in and among each plot.

RESULTS

A. cephalotes exhibited equal selection of all of the plant extracts within the Shade-Grown Plot (Table 2). The same was true within the Monoculture Plot and the No-Coffee Plot (Table 2).

TABLE 2. Kruskal-Wallis values show no difference for *A. cephalotes* preference of three plots on coffee farms, evaluated for six plant extracts (*E. lanceolata*, *S. glandulosum*, *C. costaricensis*, *A. arborescens*, *C. arabica* [Caturra], and *C. arabica* [Katway]) and a control extract, in Cañitas, Costa Rica.

	df	n	Kruskal-Wallis value	p-value
Shade-Grown Plot	6	35	0.976	0.98
Monoculture Plot	6	35	1.319	0.971
No-Coffee Plot	6	35	0.803	0.992

The ants selected *E. lanceolata* in the Shade-Grown Plot more frequently than in the other sites (daily average = 67 oats; Kruskal-Wallis = 7.22, df = 2, $p = 0.027$, $n = 15$; Fig. 1, Table 3), followed by the No-Coffee Plot (average daily removal = 48 oats), and the Monoculture Plot removed the fewest *E. lanceolata* oats (daily average = 29).

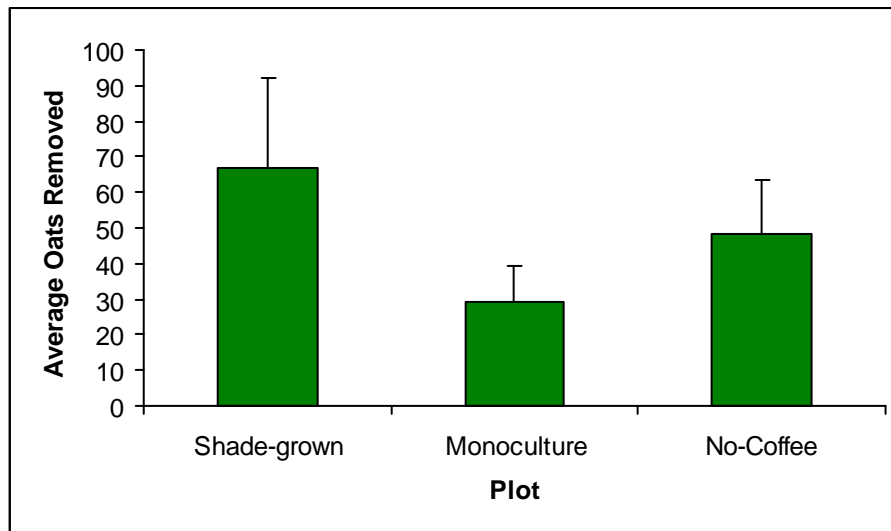


FIGURE 1. Difference in average daily removal of oats with *E. lanceolata* extract by *A. cephalotes* at three different plots (Kruskal-Wallis = 7.22, df = 2, $p = 0.027$, $n = 15$) in Cañitas, Costa Rica.

A. cephalotes showed a trend toward selecting *S. glandulosum* more frequently at the Shade-Grown Plot than the other plots (Table 3). The ants did not show a preference among the plots with their selections of *A. arborescens*, *C. costaricensis*, Caturra, or Katway (Table 3).

TABLE 3. Kruskal-Wallis values for *A. cephalotes* selection of oats of six plant extracts (*E. lanceolata*, *S. glandulosum*, *C. costaricensis*, *A. arborescens*, *C. arabica* [Caturra], and *C. arabica* [Katway]) and a control extract among three plots on coffee farms in Cañitas, Costa Rica. Shade-Grown Plot grows shade coffee (*C. arabica*) of Caturra and Katway varieties; Monoculture Plot grows both varieties in monoculture; No-Coffee Plot is a field of tall grass with no coffee. Each of the three plots has a separate *A. cephalotes* colony.

	df	n	Kruskal-Wallis value	p-value
Control	2	15	1.86	0.395
Caturra (<i>C. arabica</i>)	2	15	0.98	0.613
Katway (<i>C. arabica</i>)	2	15	1.52	0.468
<i>Erythrina lanceolata</i>	2	15	7.22	0.027
<i>Sapium glandulosum</i>	2	15	5.58	0.061
<i>Citharexylum costaricensis</i>	2	15	2.56	0.278
<i>Acnistus arborescens</i>	2	15	1.22	0.543

DISCUSSION

These results did not support the hypothesis that *A. cephalotes* would select more frequently for the coffee varieties than the shade tree species. Instead, these results suggest that designing shade-grown may be beneficial for reducing herbivory on coffee leaves in Cañitas, Costa Rica. *A. cephalotes* did not display a preference for one or more of the shade tree species over the two varieties of *C. arabica*, but it did select all of the species equally at each plot. Therefore, planting these shade species among coffee plants would provide multiple targets for the ants, alleviating the harvesting of coffee leaves.

It has been shown that extracts from fast-expanding young leaves in the Tropics are preferred for feeding and also support greater fungal growth (Coley and Barone 1996). Also, the concentrations of tannins, carbon-based metabolites in leaves that are used for defense against herbivory, are known to vary based on available nutrients and light (Denslow et al. 1987, Mole et al. 1988) during leaf maturation. Therefore, there

may be certain ages or growth conditions at which ants would selectively cut these shade species, in turn favoring coffee.

Additionally, these results show that *A. cephalotes* more frequently select oats of the *E. lanceolata* extract at the Monoculture Plot in the shade-grown coffee. This may offer particular benefits to farmers who already practice traditional coffee growing. *E. lanceolata* could be especially useful as a shade tree within an already shaded plot or plantation to reduce leaf herbivory. In addition, farmers already frequently plant and prune nitrogen-fixing legumes as a method of soil nitrification (McGrath 2000), making *E. lanceolata* an even better option for a shade-grown plantation. The ants may even be familiar with this species as it has been used before for nitrification on coffee farms.

E. lanceolata, *S. glandulosum*, *C. costaricensis*, and *A. arborescens* are all known as fast-growing species, which need little maintenance, and grow well in disturbed areas. Many of these species also can propagate from cuttings (Stevens 1983), which minimizes time to maturity. Therefore, the easy fast establishment of these species (Woolliams 1979) allows them to establish early in the typical, eight-to-ten-year life of a coffee crop (Stevens 1983). Fresh *Erythrina* seeds, up to three-months-old, often have up to 100% germination rates when sown (Woolliams 1979). Cuttings from *A. arborescens* can be grown as living fence posts, serving multiple purposes as coffee shade, fence post, and a target for *A. cephalotes* (Haber et al. 2000).

From an ecological standpoint, it is particularly exciting that equal selectivity of the tested plant species by *A. cephalotes* was observed. For example, all 112 species of *Erythrina* in the tropics and subtropics are adapted for bird pollination, by either passerine birds or hummingbirds (Neill 1988). Wasps visit *S. glandulosum*, while small moths and stingless bees pollinate *C. costaricensis* (Haber et al. 2000). *A. arborescens* attracts bees, beetles, butterflies, and occasionally hummingbirds for pollination (Haber et al. 2000). The addition of even these few species of shade trees to a monoculture or shade-grown coffee plantation has great potential to allow the establishment of numerous animal populations to an area with low biodiversity.

A. cephalotes is also known to cut non-leaf plant parts, including flower buds. During this study, I observed ants at all of the plots carrying large quantities of multiple, unidentified flower buds to the nests. A further study could examine *A. cephalotes* selection among coffee flower buds and the buds of a collection of shade species to possibly strengthen the importance of shade-grown coffee for both farmers and biodiversity. Leaf toughness is the most effective defense against herbivory (Coley and Barone 1996). Using leaf fragments, as opposed to oats, in a similar study could provide valuable information about how planting shade tree species with varying leaf toughnesses would affect leaf herbivory of coffee. Also, multiple farmers and residents of the plots stressed the high amounts of damage that *A. cephalotes* does to both roses and *Heliconia*, so either of these plants may offer additional studies in Cañitas, Costa Rica. Finally, a study could look at leaf-cutter ant damage to coffee plants in monocultures versus shade-grown coffee plants.

ACKNOWLEDGEMENTS

I would like to thank Tania Chavarría for her suggestions in refining my methods, and for her assistance with analyzing the results of the study. I would also like to thank Karen Masters for her assistance in selecting appropriate shade tree species, as well as helping to familiarize me with them. Also, thank you to Richard Fritzmeier for help with locating two of my research sites. Thanks to Juan Cruz Jiménez and Victor Torres Rojas for allowing me access to their coffee plantations. Finally, thanks to Josh Cooper for reviewing this paper.

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