Causes and consequences of diversity, complexity, and autonomous ecosystem services in coffee agriculture

by

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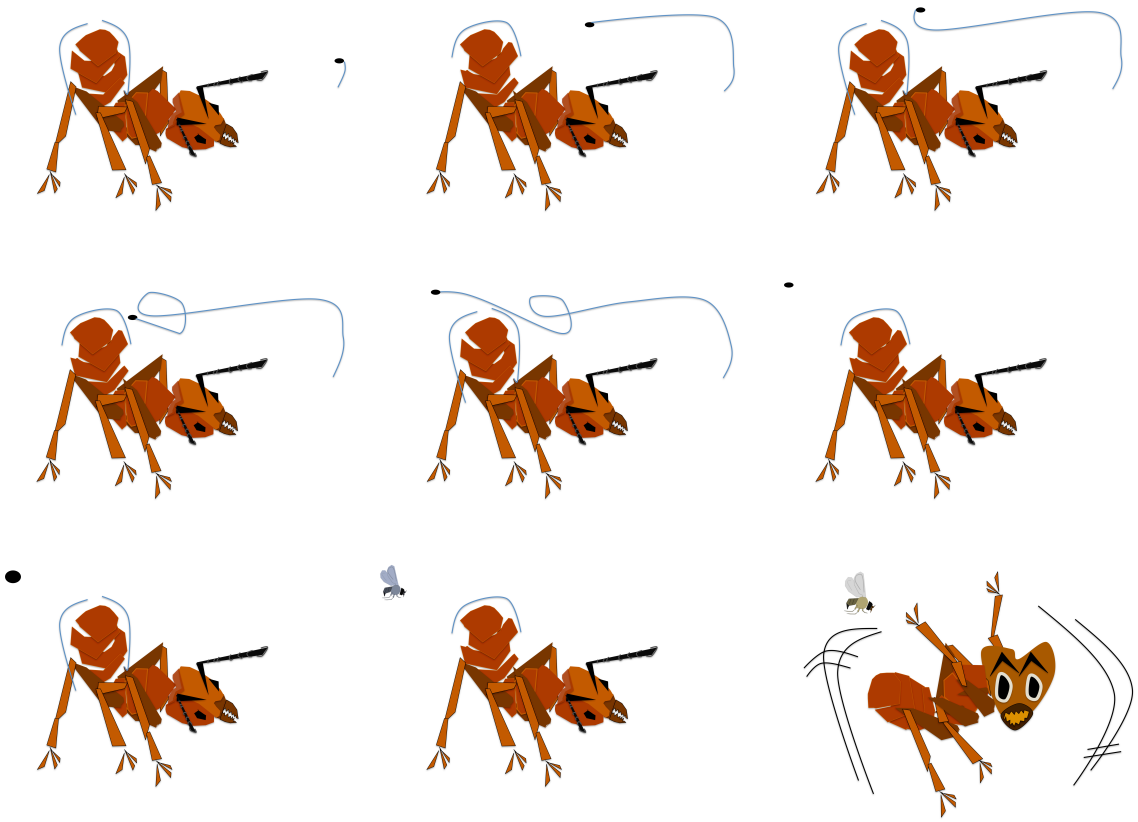

Non-consumptive effect of the decapitating fly on ant host

Dedication

To Peter, Connie, and John Gonthier.

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Abstract

Agricultural production systems, like other land-use types, are made up of ecological communities that contribute to ecosystem processes. The importance of biological complexity in the form of diversity, food chain length, and non-trophic interactions is not often recognized as an important component in agricultural systems. This lack of recognition persists even though in many other fields, from economics to medicine, embracing complexity has revealed important insights into better practices. Current agricultural practices frequently simplify ecological communities to make way for mechanization and the maximization of crop yield in monoculture. But these conventional methods in agriculture produce many novel problems associated with soil processes, pollination, and pest and disease outbreaks. Considering the ecology of the agricultural ecosystem may help us to address many of these problems, even in the most-simple monoculture. This dissertation addresses two over-arching questions related to ecological complexity and agriculture: What factors are important for maintaining biodiversity within agricultural systems? And, what ecosystem services are lost following agricultural intensification and/or the loss of species?

Chapter 1 introduces the problem of biological complexity in agricultural systems. Chapter two uses a quantitative synthesis of the literature to show that agriculture impacts species richness and abundance at local management and landscape scales. Further, to conserve species in agriculture, policy programs need to take a local and landscape level strategy, as some groups of species only respond to effects at a single spatial scale. In chapter three, the evaluation of stable isotope analysis, an indirect measure trophic feeding position, shows that intensification can shift trophic position, limit trophic diet breadth, and modify the overlap in the resources used between species. Chapter four provides evidence that six of eight ant species studied suppress an important coffee pest from colonizing harvestable fruits. Chapter five shows that multiple predator species suppress diverse pest communities better than any single predator species. However, single predator species are equivalent to multiple predator species at suppressing individual pest species. These results suggest that focusing on the effects of multiple predator

species on entire pest communities may better reveal a greater importance of predator biodiversity. In chapter six, observational and experimental studies show that the strength of an interaction cascade is context dependent and results in a different suite of services being provided to crop production. Chapter seven synthesizes the results of this dissertation and reflects on their significance to agriculture.

This dissertation points to small and large scale effects of agriculture on biodiversity that have clear policy implications for conservation in agricultural landscapes. The dissertation also outlines other modifications to the niche ecology of species in intensified and non-intensified systems. Further, the work shows that to achieve holistic pest suppression services, multiple predator species are needed to suppress entire pest communities. Finally, the results suggest that complex interaction cascades are context dependent and may result in the delivery of different suites of services in agricultural systems. Thus, this dissertation provides a case study of the causes and consequences of biodiversity loss in agriculture and highlights the importance of considering biological complexity for the maintenance of autonomous ecosystem services.

Chapter 1
Introduction

While complexity may sometimes make it difficult to understand the effect of one subject on another subject (Yodzis 2000), advances in network analysis suggest that considering additional existing links within networks increases the predictability of one-on-one interactions (Berlow et al. 2009). Yet, in many fields the predominant line of research focuses on the minimal components (i.e. the focal interaction) for reasons of feasibility or because there is an implicit assumption that distant indirect interactions between nodes, which are connected by long chains, are probably weak and therefore unimportant (Berlow et al. 2009). In the field of ecology, a central goal is often to determine the importance of complexity in maintaining the functioning of systems. In particular, ecologists seek to understand the how environmental factors influence the number of species, abundance, evenness, composition, other food web metrics of complexity (e.g. linkage density, compartmentalization, etc.), and non-trophic interactions (e.g. mutualistic interactions, behavioral modifications, etc.) and the resulting consequences of these changes for ecosystems (Duffy et al. 2007, Tylianakis et al. 2007, Berlow et al. 2009, Cardinale et al. 2012). Further, the abiotic, anthropogenic, and other ecological factors that modify complexity are now being described to help draw connections between the degradation of ecosystem services and the loss of diversity and ecological complexity.

There are a growing number of examples in the economic, social, and medical sciences that have endorsed the importance of some aspects of complexity. Perhaps one of the most pronounced examples of the use of complexity in terms of diversity comes from financial investments where diverse relative to simple portfolios tend to reduce the risk of investment failure. Although not universal, diversification is widely held to be a key tenet of modern portfolio management (Shawky and Smith 2005). More recently the health and medical sciences have moved to try to understand the positive and negative impacts of microbial organisms living on and within the human body (Peterson et al. 2009). The National Institute of Health's Human Microbiome

Project (HMP) is a major undertaking with a planned investment of $150 million over 5-years (Peterson et al. 2009). One of the main observations leading to this huge project was a reassessment of the assumption that all microbial organisms living in the human body are detrimental to human health. In fact, many microbial organisms may provide direct or indirect benefits within the human-ecosystem. There are already striking examples of the importance of this complexity. For instance, recurrent gut infections by *Clostridium difficile* (pathogenic bacteria) are best treated by reestablishment of the gut micro-fauna from a healthy donor through fecal transfer to an infected individual. Reestablishing a diverse micro-fauna had an 81% success after one infusion, which was a far greater treatment relative to the 31% success rate of traditional anti-biotic treatments (van Nood et al. 2013). Thus, it appears one of the great insights to be gained from these HMP studies will reveal the negative consequences of sterilizing our human-ecosystem of its microbiome through the overuse of antibiotics.

Falling behind the health sciences, the agricultural sciences still embrace a vast number of practices that have drastic effects on the ecological components of agricultural ecosystems. This rapid transformation over the last years has been astounding. A number of technological advances have enabled the mechanized production and harvest of crops, but the need for high organization has resulted in monoculture planting of crops of just a few varieties (Altieri 1987, Vandermeer 2010). Similarly, the reliance on chemical fertilizers has bypassed the short-term need to maintain and build complex organic matter in the soil (Howard 1940). Further, the introduction of synthetic pesticides (targeting microbes, plants, and animals) has caused additional simplification of the ecological communities within fields (Lewis et al. 1997). The increasing size of farms, and total area covered in farmland has also resulted in less natural habitat between agricultural fields (Tscharntke et al. 2005).

What are the consequences of this ecosystem simplification? A great body of literature has documented how the many steps of intensification across many crops has resulted in the decline in species abundance, richness, and many changes to community composition and network structure (Bengtsson et al. 2005, Tscharntke et al. 2005, Tylianakis et al. 2007, Attwood et al. 2008, Philpott et al. 2008, Macfadyen et al. 2009, Batáry et al. 2011, Macfadyen et al. 2011). Agriculture's impact on ecological communities is complex and is manifested at small field

scales and also in larger landscape, regional, and global patterns (Tscharntke et al. 2005). At the same time, the ecological literature suggests that less diverse ecological communities are, on average, less efficient at maintaining ecosystem functions (Cardinale et al. 2012). This suggests that agricultural simplification has the potential to drive both environmental and production problems. Indeed, some agricultural research now describes how simplification of communities may result in the loss of important natural limiting agents of a number of agricultural problems related to productivity and agricultural pollution (Lewis et al. 1997, Klein et al. 2003, Vandermeer et al. 2010).

This dissertation is a case study into the effects of agricultural intensification on biodiversity and ecological complexity, as well as the consequences of those community components on services delivered to agriculture, with an emphasis on pest control. Chapter 2 takes a broad look at the maintenance of species diversity in agricultural systems. It uses meta-analytical techniques to describe how diversity in different taxonomic groups responds to both small- and large-scale agricultural effects. Chapters 3-5 focus on coffee agroforestry systems that are well recognized for their great variation in tree diversity and food web complexity. Chapter 3 specifically asks how coffee management for high and low vegetation complexity alters the trophic resource use of four ant species. It takes advantage of stable isotopes as a indirect measure of trophic resource use to determine if management intensification shifts the trophic position of species, limits the diversity of trophic resources consumed (trophic niche width), and alters the overlap in trophic niches between species. Chapter 4 investigates the importance of ant pest suppression of a major pest of coffee, the coffee berry borer. It discovers that six of eight ant species studied suppressed the colonization of coffee berries by the borer. Chapter 5 aims to uncover any synergistic effects of ant diversity on pest suppression. It investigates whether incorporating single pest species or multiple pest species in treatments modifies the importance of multiple ant predator species. Chapter 6 investigates how an interaction web, including a dominant ant species, coffee, and three pest (insects and a pathogen) species, is modified by the species identity of the nest tree that hosts the dominant ant species. This study builds from previous research at the site that has long described this interaction web and suggests that the nest tree species identity alters the strength of interactions between species resulting in trade-offs in ecosystem services. Finally, chapter 7 provides a final synthesis of these projects.

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Chapter 2
Biodiversity conservation in agriculture requires a multi-scale approach

Abstract

Biodiversity loss - one of the most prominent forms of modern environmental change - has been heavily driven by terrestrial habitat loss and, in particular, the spread and intensification of agriculture. Expanding agricultural land-use has led to a prominent debate, with some arguing that biodiversity conservation in agriculture is best maximized by reducing local management intensity, like fertilizer and pesticide application. Others argue that landscape level approaches that incorporate natural or semi-natural areas in landscapes surrounding farms are needed. Here we show that both sides of this debate are partially correct, and that either local or landscape factors can be most crucial to conservation planning depending on which types of organisms one wishes to save. We performed a quantitative review of 266 observations taken from 31 studies that compared the impacts of localized (within farm) management strategies and landscape complexity (around farms) on the richness and abundance of plant, invertebrate, and vertebrate species in agro-ecosystems. While both factors significantly impacted species richness, the richness of sessile plants was solely associated with local management effects, whereas the richness of more mobile vertebrates was only associated with landscape complexity. Invertebrate richness and abundance responded to both factors. Our analyses point to clear differences in how various groups of organisms respond to differing scales of management, and suggest that preservation of multiple taxonomic groups will require multiple scales of conservation.

Introduction

One of society's most pressing challenges is to slow the rate of global biodiversity loss and extinction (Vitousek et al.1997; Loreau et al.2001, Tilman et al. 2001). There is now

overwhelming evidence that the loss of species impacts the functioning of ecosystems (Cardinale et al. 2012) and that many services provided by species have important economic value (Daily 1997). Much conservation research has therefore focused on where biodiversity is being lost most rapidly and where the loss of biodiversity will have the most immediate consequences. Of the drivers of global biodiversity loss, the widespread conversion of land to monoculture crop production, and the intensification of local agricultural practices, such as through fertilizer and pesticide use, is considered to be among the most damaging to biodiversity (Sala et al. 2000, Donald et al. 2001, Green et al. 2005, Tscharntke et al. 2005, Perrings et al. 2006, Attwood et al. 2008, Perfecto et al. 2009, Clough et al. 2011, Phalan et al. 2011). In turn, the services that species provide related to pest control, pollination, and nutrient cycling that benefit agricultural production and sustainability could be compromised (Tscharntke et al. 2005). With ever increasing global demands for agricultural production of food and fuel, additional stresses on species in and surrounding agricultural land are inevitable (Tilman et al. 2001). As such, the conservation of biodiversity living in agriculture has become a major focus of much conservation policy.

But how we best conserve biodiversity in agricultural fields remains a contentious debate. Programs in numerous countries have attempted to reduce the severity of agriculture's negative influence on biodiversity by paying farmers to reduce management intensity through reduced pesticide inputs, synthetic fertilizer inputs, or by converting farms to organic practices (Kleijn & Sutherland 2003, Bengtsson et al. 2005, Attwood et al. 2008, Kleijn et al. 2011, Whittingham 2011). Several syntheses suggest that reduction in local management intensity does conserve biodiversity (Bengtsson et al. 2005, Attwood et al. 2008), but other empirical research has failed to support these claims (Kleijn et al. 2001, Kleijn & Sutherland 2003, Kleijn et al. 2006). Proponents of meta-population and meta-community theory are quick to point out that reduced intensity of one small farm may do little to conserve species with large range sizes or species that require adjoining subpopulations in the surrounding landscape (Tscharntke et al. 2005, McKenzie et al. 2013). Furthermore, reduced intensity may provide little benefit if a farm is surrounded by a landscape of high-input intensive farming because poor species pools in the desolate landscapes may limit the colonization of the wildlife friendly farm (Ricketts et al. 2001, Tscharntke et al. 2005). These critics propose that agriculture's larger-scale effects, the

homogenization of entire landscapes (Meehan et al. 2011), may be the primary factor driving biodiversity loss (Ricketts et al. 2001, Tscharntke et al. 2005, Batáry et al. 2011, Chaplin-Kramer et al. 2011, Concepción et al. 2012). In order to conserve biodiversity in agriculture, they argue, we need to incorporate more natural and semi-natural habitats in areas surrounding farms and/or maintain high habitat diversity in agricultural landscapes. Thus, one conservation strategy targets local management intensity and the other targets larger scale management of landscapes (Gabriel et al. 2010, Whittingham 2011, Kennedy et al. 2013).

Given that species can vary in many functional traits, such as, mobility, range size, dispersal capability, and sensitivity to agricultural activities, biodiversity in different taxonomic groups may respond to different scales of agricultural intensification (Tscharntke et al. 2005, McKenzie et al. 2013). For example, while some plants may have high seed-dispersal capability, they are all sedentary organisms. Therefore the application of herbicide within farms may largely eliminate many species from farmlands. Other organisms, like mammals and birds, are capable of foraging across many habitats and over a large spatial area. These species may require landscape level features to persist in farmlands. Although some empirical studies have supported the hypothesis that multiple scale factors limit biodiversity in agriculture (Gabriel et al. 2010, McKenzie et al. 2013), a broad synthesis of the published literature is lacking.

We performed a quantitative review to investigate the influence of local management intensification and landscape complexity on biodiversity in agriculture. We reviewed 31 field studies that provided 266 observations of how the species richness and abundance of plants, invertebrate, and vertebrate animals differed between agro-ecosystems with low- versus high-local management intensities, and varied with surrounding landscape complexity. Low-intensity agro-ecosystems consisted of farms that were certified organic or had reduced chemical inputs or reduced planting and/or grazer densities as compared to high-intensity conventional agro-ecosystems (see Methods). Landscape complexity was defined as the proportion of natural and semi-natural areas (non-crop lands) or the variety of different habitat types (measured as Shannon's Diversity Index) in landscapes surrounding farms. We predicted that sessile organisms like plants would be influenced more by local than by landscape strategies due to their low mobility that makes them susceptible to disturbances at small spatial scales. We predicted

that more mobile organisms would be buffered against local management intensification, and respond more to landscape-scale management. As we show next, both local and landscape strategies are needed to promote plant, invertebrate, and vertebrate biodiversity because different groups respond to different scales.

Methods

Selection of studies

We conducted an ISI Web of Science literature search of studies that compared species richness and abundance in low- and high-intensity agricultural fields that were nested within a gradient of landscape complexity (last search January 13, 2012). In addition to our primary search, we also reviewed the reference sections of several recent reviews and meta-analyses (Tscharntke et al. 2005, Batáry et al. 2011, Chaplin-Kramer et al. 2011) and obtained papers through author contacts (Jonason et al. 2011, Jonason et al. 2013). In total, we reviewed 822 published studies. Inclusion of a study within our quantitative review was contingent on the following criteria: 1) the study must have collected empirical data on species richness within agriculture. 2) The study must have compared categorically defined local-scale factors related to agricultural management intensity. 3) The study must have included variation in landscape-scale factors related to landscape complexity in each sampling site. These measurements must have been taken at a minimum scale of 1.96ha (~250m radius surrounding the sampling site).

Of the 822 published studies reviewed, 44 fit our criteria (33 from search, 11 from references). Some studies reported results from the same datasets; see references (Weibull et al. 2000, Weibull et al. 2003) and references (Geiger et al. 2010, Fischer et al. 2011, Flohre et al. 2011). We were unable to obtain the data from 9 studies. In the end, we obtained data from 31 studies (Weibull et al. 2000, Letourneau and Goldstein 2001, Weibull et al. 2003, Clough et al. 2005, Purtauf et al. 2005, Roschewitz et al. 2005, Schmidt et al. 2005, Batáry et al. 2006, Gabriel et al. 2006, Holzschuh et al. 2006, Rundlöf and Smith 2006, Batáry et al. 2007, Rundlöf et al. 2007, Sjödin et al. 2007, Winfree et al. 2007, Batáry et al. 2008, Haenke et al. 2009, Batáry et al. 2010, Culman et al. 2010, Dänhardt et al. 2010, Geiger et al. 2010, Holzschuh et al. 2010, Smith et al. 2010, Fischer et al. 2011, Flohre et al. 2011, Jonason et al. 2011, Kovács-Hostyánszki et al. 2011, Winqvist et al. 2011, Batáry et al. 2012, Jonason et al. 2013). We were unable to recover all data

from some highly collaborative studies that spanned multiple countries and research groups because of data-sharing issues across the large-scale projects (Geiger et al. 2010, Flohre et al. 2011, Winqvist et al. 2011). For richness, we obtained a total of 70 observations for local factors and 71 observations for landscape factors (from 31 studies). We obtained observations of abundance, activity abundance, or percent cover for a total of 62 observations for local factors and 63 observations for landscape factors (28 studies) (see Fig. 2 for taxonomic group sample sizes). Two outliers were removed from the dataset for analysis of local management statistical models to improve the model fit and the normality of the data; for plant richness (Flohre et al. 2011) and plant abundance (Batáry et al. 2012).

Local management factors

All local scale management factors fell under a comparison of a low-intensive form of agricultural practice versus a high-intensive form of agricultural practice. Low-intensity agriculture consisted of certified organic practices, practices in compliance with an agri-environment scheme aimed to benefit the environment or biodiversity (i.e., the planting of flower strips in field margins); see ref. (Kleijn & Sutherland 2003), and extensified agriculture in which chemical inputs, plant densities, or grazing densities were low. High-intensity farms had conventional management levels of chemical inputs and planting or grazing densities that were always greater than the low-intensity farms they were compared against in each study. For each measurement of species richness or abundance (y) within a study, we calculated the local management effect size on biodiversity as a log response ratio $LR_M = ln(y_L/y_H)$ where y_L is the mean of biodiversity in low-intensity farms and y_H is the mean of biodiversity in high-intensity farms. Log response ratios are unitless metrics that allow us to determine if there is a proportional difference between mean levels of species richness in low- and high-intensity farms (Hedges et al. 1999, Borenstein et al. 2009). In studies from Sweden, study designs were such that low- and high-intensity farms were paired to control for variation in management type and location (Weibull et al. 2000, Rundlöf & Smith 2006, Rundlöf et al. 2007, Dänhardt et al. 2010; Smith et al. 2010). Log response ratios compare un-paired means between low and high intensity farms, therefore for these Swedish studies, the effect sizes calculated had less power than if we were able to maintain a paired design within our analysis.

Landscape factors

We obtained 170 correlations between species richness and landscape factors and 161 correlations between total abundance and landscape factors. We excluded all measurements less than an area of 1.96 ha (the area of a circle with a radius of 250m). We excluded measurements of mean field size, length of habitat boundaries, percent grasslands, and percent intensive agricultural area because they do not intuitively correlate with landscape complexity. In one case, we included a measure of percent grassland because authors stated clearly that it was strongly positively correlated with the diversity of habitat types (Purtauf et al. 2005). We defined the landscape factors percent natural area, semi-natural area, and woodlands as percent non-crop area. We also defined the inverted percentage of arable land, croplands, managed lands and agriculture as the percent of non-crop area and assumed all measures of non-crop area correlated with landscape complexity. We also included the diversity of habitat types (measured as the Shannon's Index) in the analysis even though it is measured on a different range of values (0 to ∞) than the percentage non-crop area. Both the diversity of habitat types and the percent non-crop area are considered important components of landscape complexity across the literature sampled (Tscharntke et al. 2005). For landscape factors, we calculated correlation coefficients (R) that related y (richness or abundance) to the measure of landscape complexity, and then standardized the coefficients to Fisher's Z as: $Z_L = 0.5 \times ln(1+R_L)/(1-R_L)$ (Borenstein et al. 2009) where Z_L is Fisher's Z and R_L is the correlation coefficient of y versus landscape complexity. Quantitative reviews comparing continuous variables often use R or Fisher's Z as an effect size because they are intuitively interpreted and they are standardized to take into account the original scales of different metrics (Borenstein et al. 2009).

Analysis

We adjusted the analysis of this synthetic review to take into consideration a number of within and between study non-independences. To account for the fact that some studies had multiple observations, we created a block by study (random effect of study). For studies with repeated measures of richness or abundance across year or season (Roschewitz et al. 2005, Schmidt et al. 2005, Holzschuh et al. 2010), we summed means and standard errors before calculating local management LR_M and averaged across the repeated measures for the landscape factor Z_L. To account for studies investigating different cropping types, we created a fixed effect of cropping type (cereal, mixed, vegetable/fruit, and pasture/meadow). Observations also varied by

geographic location and by research group (affiliation). Therefore, we created a random effect of country that accounted for these non-independences to a large degree. To account for the differences between landscape factors, we included a fixed effect of landscape factor type (percentage non-crop area or habitat diversity) and the continuous covariate, landscape scale.

We performed general linear mixed models (GLMM) to determine if mean local management (LR_M) and landscape (Z_L) effect sizes differed from 0 and to compare the differences between taxonomic groups (West et al. 2007). For all GLMMs, we performed type III F-tests of significance for main effects with maximum likelihood to estimate the fixed effect parameters and variance of the random effects. For each GLMM, we included the random effect of study and determined if the random effect of country improved model fit. However the random effect of country generally had a very small estimated variance and often resulted in problems related to over-fitting the model, therefore we proceeded without this factor within models. For the local management level models, we used the response variable LR_M and entered taxonomic group and crop type into the model as fixed effects and the study as a random effect. For landscape level models, we used the response variable Z_L and entered taxonomic group, crop type, landscape factor type, and landscape factor scale (covariate) as fixed effects and a random effect of study. We performed model selection using Likelihood Ratio Tests to exclude fixed effects that did not improve model fit (West et al. 2007). We used the final GLMM models to estimate mean and 95% confidence intervals of each effect size with the function EMMEANS in SPSS (20.0). Mean effect sizes that were significantly more positive or more negative than 0 were interpreted as significant at $\alpha = 0.05$. In addition to un-weighted effect sizes, we also ran analyses with effect sizes weighted by the inverse of the variance (Hedges et al. 1999). We present un-weighted models because discrepancies between un-weighted and weighted models were small, and un-weighted models allow observations with few large plots to have the same effect as observations with many small plots. We conducted all statistical analysis in SPSS (20.0).

Failure to publish negative or non-significant results with low samples size can result in literature for which outcomes are biased and strongly positive. Therefore in quantitative syntheses it has become commonplace to test for the importance of publication bias using a number of methods. If a correlation between sample size and effect size exists many argue this is evidence for bias

toward publication of studies with positive effects with large sample sizes. Failsafe numbers are also used to estimate the number of studies with null results needed to eliminate the significance of a statistical analysis. We calculated Rosenthal's fail-safe number and correlated sample size versus effect size to evaluate the importance of any potential publication bias.

Results

Our results consistently show that both local management and landscape complexity impact species richness (Fig. 2.1A, Table 2.1). However, the importance of each factor differs among the three taxonomic groups examined (Fig. 2.2A,C). Overall, 52 out of 70 (74%) observations showed that low-intensity farms had more species than high-intensity farms (Fig. 2.1A). Mean overall richness, estimated across all organisms, was 40% higher in low-intensity relative to high-intensity farms (Fig. 2.2A). While plant and invertebrate richness was 92% and 21% higher in low-intensity relative to high-intensity farms, respectively, vertebrate richness did not differ significantly among local management types.

Forty-seven out of 71 (66%) observations showed a positive relationship between landscape complexity and species richness within farms (Fig. 2.2C). The mean correlation between plant richness and landscape complexity was not significantly positive (Fig. 2.2C), suggesting that landscape factors were less important to plant biodiversity than were local management factors. In contrast, both invertebrate and vertebrate animals had significantly positive mean correlations, indicating that species richness of these groups increased as a function of increasing landscape complexity. For vertebrates, which showed no association with local management intensity, this result suggests landscape complexity is more important than local management factors.

The analysis of taxonomic abundance also revealed important patterns. For the local management scale, overall 44 of 63 (69.8%) observations found higher abundance in low-intensity relative to high-intensity farms (Table 2.2, Fig. 2.1B). Although the mean overall abundance, estimated across all organisms, was 27% higher in low-intensity relative to high-intensity farms, within taxonomic groups, vertebrate and plant abundance did not different between the two local management types (Fig. 2.2B). Only invertebrate abundance was significantly greater in low- relative to high-intensity farming.

The overall mean correlation between abundance and landscape complexity was significantly positive, with 36 of 64 (56.2%) observations positively correlated (Table 2.2, Fig. 2.1B). However, within taxonomic group, only invertebrate abundance was significantly positively correlated with landscape complexity (Fig. 2.2D). For invertebrates, this suggests that the higher levels of richness in systems that are farmed less intensively, or that are surrounded by more complex habitats, could be driven by an increase in the overall abundance of invertebrates (it is well-known that the discovery of species is proportional to the number of individuals sampled). In contrast, the abundance of plants and vertebrates did not vary with local or landscape factors, which suggests that higher levels of richness were independent of any impacts of factors on population sizes for these two groups (Fig. 2.2).

Across our observations there were no significant correlations between effect size and n for management LR_M or landscape Z_L and for richness and abundance data (Table 2.3). Rosenthal's fail-safe analysis suggested that at least 100 nil observations were needed to eliminate statistical significance ($p < 0.05$) across all analyses with significant mean effect sizes, except in one case. The fail-safe number for the landscape Z_L of vertebrate richness was only 32.5, therefore we caution the interpretation of this result, but note that 32.5 nil observations are still 3.6 times more observations than the number of existing observations.

Discussion

Conservation strategies in agriculture have focused on reductions in local management intensity and on increasing the amount of natural area or habitat diversity in agricultural landscapes. Our results suggest that both of these strategies are needed to promote plant, invertebrate, and vertebrate biodiversity because different groups respond to different scales. The strong decline in plant richness in intensively managed farms is likely the intended outcome of agricultural practices designed to eliminate weedy crop competitors. Herbicide application, synthetic fertilization of crop plants, and tilling can have direct or indirect negative effects on plant diversity within agricultural systems (Roschewitz et al. 2005, Tscharntke et al. 2005). Though the reductions in plant diversity may well be the farmer's intention, this does not negate the significance of this loss of biodiversity. Many species considered to be arable "weeds" are

categorized as threatened on the IUCN red list (e.g., 38% of arable plants in Germany are red listed species (Tscharntke et al. 2005)) and these local management practices are strongly eliminating these arable species. What is, perhaps, more surprising is that there was no correlation between plant biodiversity and landscape complexity. Increased landscape complexity may promote plant diversity within farm through seed rain from neighboring non-crop habitats (Roschewitz et al. 2005), but if local management within farms is frequent and intensive, those seeds may never reach vegetative states. The observations included in our analysis all measured vegetative plants. Measurement of the plant biodiversity in the seed rain or the seed bank may help reveal any existing landscape effects on plants.

Another goal of intensification is to eliminate arthropod pests. Intentional spraying for arthropods might help explain lower species richness and abundance of invertebrates in more intensive farms if, and only if, the observations in our dataset were dominated by herbivorous pests of the focal crops. However, roughly 77% of the observations of invertebrate biodiversity (37/44) included in our analyses consisted of groups like bees, spiders, and carabid beetles, all of which are not generally herbivorous. In fact, these groups are often associated with important ecosystem services like pollination and natural pest control (Tscharntke et al. 2005, Chaplin-Kramer et al. 2011, Kennedy et al. 2013). Thus, our results suggest that the losses of species in agricultural plots due to local management and landscape level intensification are very often associated with the loss of beneficial invertebrate diversity. These unintended losses could have important negative impacts on ecosystem services like pollination and pest control.

The lack of a response of vertebrate biodiversity to local management, coupled with the strong response to landscape complexity, is almost certainly due to the high mobility of these taxa (e.g., mammals and birds) which allow them to experience the landscape at a larger scale and capture resources across larger areas in habitats outside of crop fields (Tscharntke et al. 2005). Greater mobility, in turn, has the potential to buffer these species from small-scale changes in local management (Tscharntke et al. 2005).

Although this study reveals clear patterns relating local and landscape level effects on biodiversity, the literature included in our review does have several limitations that should be

kept in mind. While many important metrics of biodiversity are well described including functional diversity, phylogenetic diversity, evenness, and other metrics of diversity, the body of literature describing local and landscape strategies for conservation in agriculture focuses on species richness and abundance. For that reason, our analysis was limited to richness and abundance. Vertebrates are poorly represented relative to other types of organisms in our dataset and most of the studies reviewed come from agro-ecosystems in Europe and the United States. We did find that our conclusions are relatively robust to select data deficiencies and the potential of publication bias, issues that are always a limitation of data syntheses (Table 2.3). In spite of these limitations, the data presently available clearly show that both local management and landscape scale strategies are important to conserving biodiversity in agriculture, as each scale influences a different set of species.

Our findings have major implications for conservation policies in agricultural landscapes. Policy strategies for conserving biodiversity in agriculture have historically focused on changing local management practices (Kleijn & Sutherland 2003, Kleijn et al. 2011). While these efforts are most certainly helpful for conserving certain groups of organisms, policy-makers and practitioners need to consider broader strategies that both reduce the intensity of local farming practices, and that use careful regional planning to place agro-ecosystems within heterogeneous landscapes so as to minimize the impact of farmlands on wildlife. As has been echoed by many other researchers, the best first steps may be to conserve existing complex agricultural landscapes and implement changes to local management practices in regions with little remaining wild lands (Tscharntke et al. 2005, Gabriel et al. 2010, Batáry et al. 2011, Kleijn et al. 2011, Concepción et al. 2012; Kennedy et al. 2013). Developing conservation plans at large spatial areas may present major challenges for future policy makers due to the difficulty in coordinating multiple land ownerships within the same landscape (McKenzie et al. 2013). Integrating multiple scales of conservation may also maximize the crop pollination, natural pest control, and nutrient cycling services that biodiversity provides (Loreau et al. 2001, Tscharntke et al. 2005, Perfecto et al. 2009, Chaplin-Kramer et al. 2011, Kennedy et al. 2013). Further, analysis of multi-scale conservation may reveal strong links between minimizing the impacts of the agricultural industry on biodiversity and maximizing nature's services to that industry.

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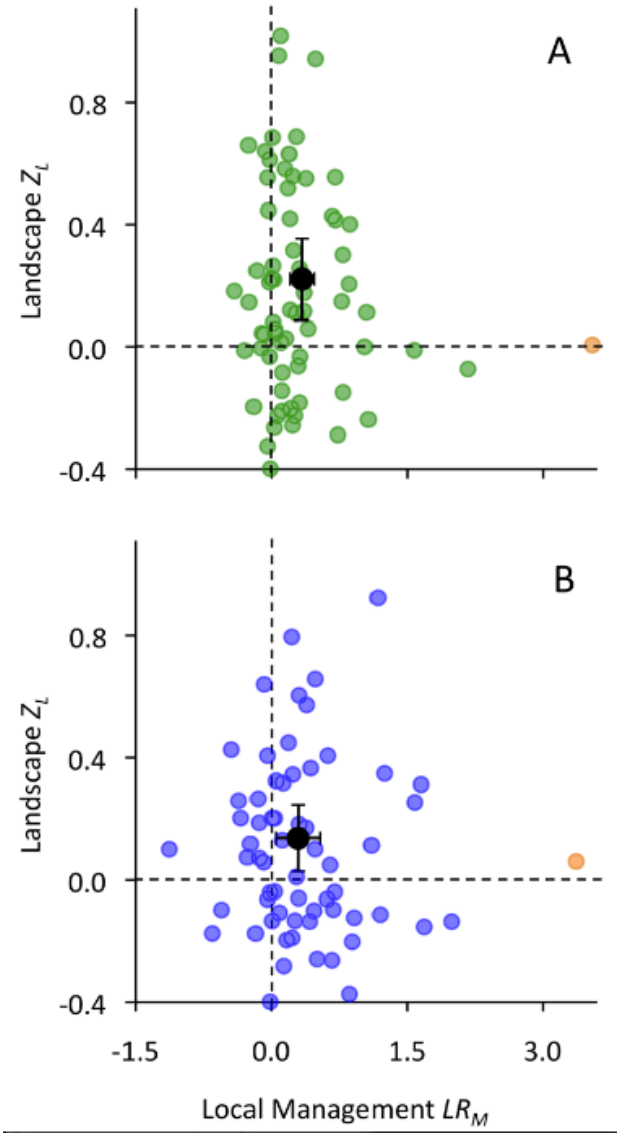

Figure. 2.1. Scatterplots of estimated marginal means and 95% CI (black points) of local management (LR_M) and landscape complexity (Z_L) effect sizes for richness (green points, n = 70 observations, 31 studies) (*A*) and abundance (*B*) (blue points, n = 62 observations, 28 studies). The x-axes represents $LR_M = ln(y_L/y_H)$, the proportional difference between mean y (richness or abundance) in low-intensity (y_L) and high-intensity (y_H) local management agriculture. The y-axes represents $Z_L = 0.5 \times ln(1+R_L)/(1-R_L)$, the transformation of R_L, the correlation coefficient calculated for y (richness or abundance) and landscape complexity. Outliers were removed from local management analysis, but remained in the landscape complexity analysis (orange points). Summary statistics of the GLMMs used to estimate marginal means and 95% CIs are available in Table 1 for richness and Table 2 for abundance.

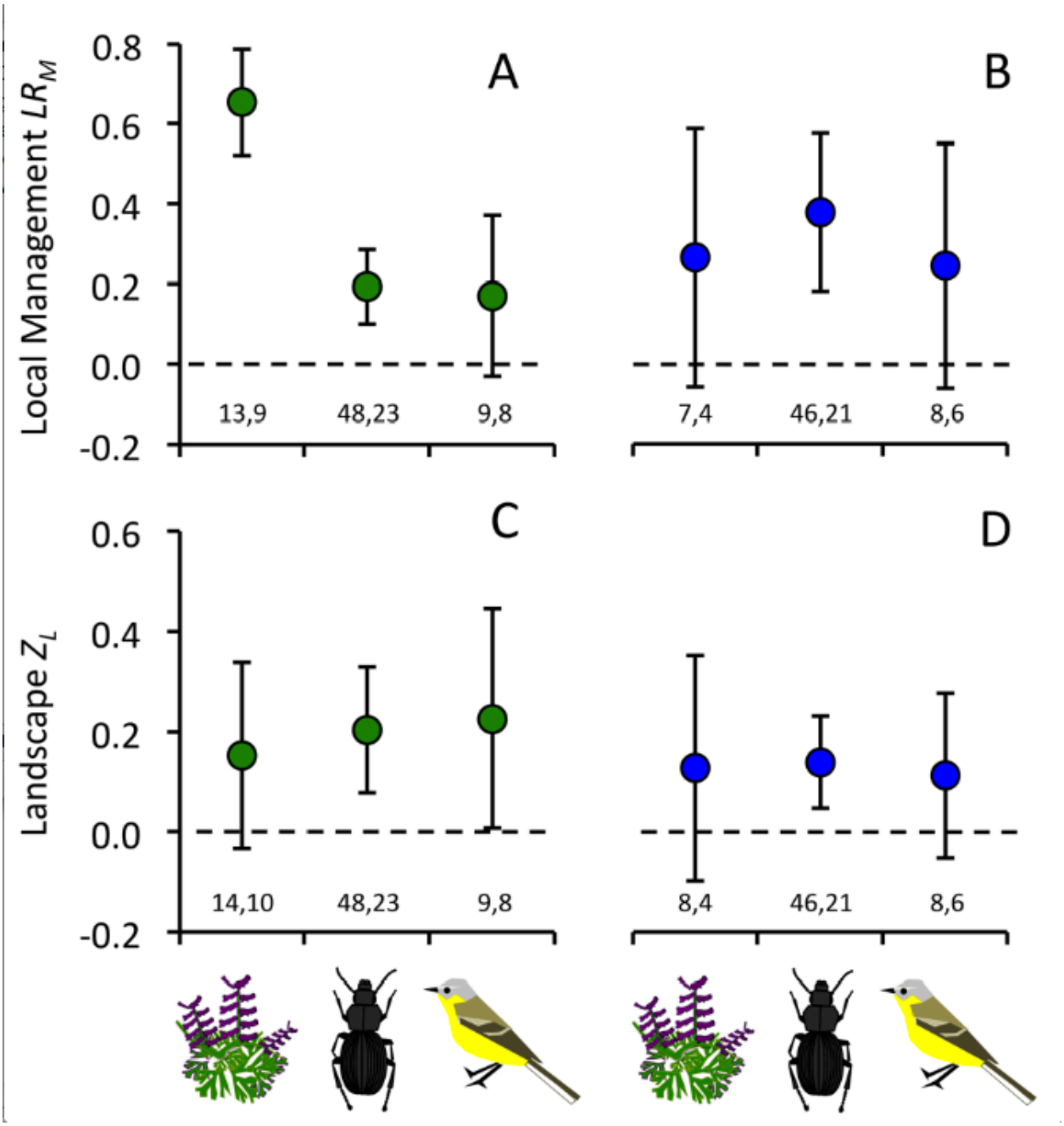

Figure 2.2. Estimated marginal means and 95% CI of plant, invertebrate, and vertebrate effect sizes for local management (LR_M) for richness (*A*) and abundance (*B*) and landscape complexity (Z_L) effect sizes for richness (*C*) and abundance (*D*). Local management $LR_M = ln(y_L/y_H)$, the proportional difference between mean *y* (richness or abundance) in low-intensity (y_L) and high-intensity (y_H) local management agriculture. Landscape $Z_L = 0.5 \times ln(1+R_L)/(1-R_L)$, the transformation of R_L which is the correlation coefficient calculated for *y* (richness or abundance) and landscape complexity. Summary statistics of the GLMMs used to estimate marginal means and 95% CIs are available in Table 1 for richness and Table 2 for abundance.

Table 2.1. Statistical models for local management and landscape complexity effect sizes on species richness. For both models a random effect of study was included. Statistical models were used to estimate mean and 95% confidence interval of effect sizes for overall responses (Fig. 2.1A) and taxonomic groups (Fig. 2.2A,C).

	*d.f.**	F	P
Local management LR_m richness			
Intercept	1,49	25.8	<0.001
Taxonomic group	2,65	8	0.001
Crop type	3,39	3.2	0.035
Landscape Z_L richness			
Intercept	1,33	10.4	0.003
Taxonomic group	2,56	0.1	0.874
Crop type	3,34	2.3	0.098

**d.f. = numerator, denominator.*

Table 2.2. Statistical models for local management and landscape complexity effect sizes on total abundance. For both models a random effect of study was included. Statistical models were used to estimate mean and 95% confidence interval of effect sizes for overall responses (Fig. 2.1B) and taxonomic groups (Fig. 2.2B,D).

	*d.f.**	*F*	*P*
Local management LR_m Abundance			
Intercept	1,30	6.1	0.020
Taxonomic group	2,55	0.3	0.756
Crop type	3,24	3.2	0.043
Landscape Z_L Abundance			
Intercept	1,63	5.1	0.027
Taxonomic group	2,63	0.3	0.722
Crop type	3,63	2.3	0.088

**d.f. = numerator, denominator.*

Table 2.3. Correlations between sample size and effect size and Rosenthal's fail-safe numbers.

	Model	Effect size vs. *n*			Fail safe	
	$P < 0.05$	*n*	*R*	*p*	*x*	*x*/*n*
Richness						
Local Management LR_M						
Overall	Yes	70	-0.130	0.284	3220.3	46.0
Plant	Yes	13	-0.157	0.610	703.2	54.1
Invertebrate	Yes	46	-0.108	0.466	627.7	13.6
Vertebrate	No	9	-0.096	0.806	12.2	1.4
Landscape Z_L						
Overall	Yes	71	-0.052	0.668	1256.9	17.7
Plant	No	14	-0.011	0.970	17.9	1.3
Invertebrate	Yes	46	-0.036	0.808	544.9	11.8
Vertebrate	Yes	9	-0.420	0.260	32.5	3.6
Abundance						
Local Management LR_M						
Overall	Yes	62	-0.226	0.076	1504.9	24.3
Plant	No	8	-0.395	0.333	0.0	0.0
Invertebrate	Yes	46	-0.196	0.191	748.8	16.3
Vertebrate	No	9	-0.116	0.767	97.0	10.8
Landscape Z_L						
Overall	Yes	63	-0.210	0.098	148.4	2.4
Plant	No	8	0.512	0.194	0.0	0.0
Invertebrate	Yes	46	-0.276	0.063	109.1	2.4
Vertebrate	No	9	-0.268	0.485	0.0	0.0

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Chapter 3
Agricultural intensification shifts, overlaps, and collapses ant trophic niche widths

Abstract

Agriculture intensification drives biodiversity loss, but its effect on other ecological processes is often un-described. Here we tested if intensification limits the range of trophic resources used by ant species (trophic niche width) and if it alters the overlap in use between species. We compared nitrogen stable isotope ratios as a relative measure of ant trophic position and niche width in coffee plantations with high and low shade-intensification. We show that intensification substantially influenced the trophic niche ecology of the ant species studied. Intensification reduced the trophic niche width of all ant species and shifted trophic position for at least one species. While intensification drove one species to a higher trophic position, other species tended to decline in position, which resulted in a change in trophic niche overlap between species. This study provides the first evidence that agricultural intensification reduces trophic niche width, alters the niche overlap between species, and is among the first to show that intensification shifts trophic position. These ecological changes may have important consequences for competitively structured communities and may help explain the loss of species following intensification.

Introduction

The conversion and intensification of agriculture are considered some of the most significant modern environmental changes to the terrestrial earth surface (Matson et al. 1997, Vitousek et al. 1997, Chamberlain et al. 2000, Sala et al. 2000, Donald et al. 2001, Tilman et al. 2001, Green et al. 2005, Tscharntke et al. 2005, Perfecto et al. 2009). One of the most alarming problems associated with these changes is the loss of biodiversity in intensified systems (Sala et al. 2000, Bengtsson et al. 2005, Tscharntke et al. 2005, Attwood et al. 2008, Perfecto et al. 2009). Many

species living in agricultural landscapes provide important services to agriculture through soil improvement, pollination, and pest control and therefore the loss of these species can have economic consequences (Daily 1997). However, how species are lost is not always clear because many other ecological changes occur during intensification (Matson et al. 1997). Very few studies track the response of competitive hierarchies, predator-prey interactions, mutualisms, and resource use by species following intensification. Intensification often involves the elimination of non-crop plant species, the planting of fewer crop species, the application of pesticides, fertilization, irrigation, and an overall simplification of the agroecosystem (Vandermeer et al. 1998, Tscharntke et al. 2005). Many organisms may lose essential resources or resource diversity during intensification (with the exception of crop pests), which may be needed to maintain viable populations within intensified systems. Reduced resource availability may also cause shifts in resource use (Benton et al. 2002, Potts et al. 2010), which may result in changes in the overlap in resource use between species. For communities dominated by competition, changes in niche overlap are thought to be very important (Hölldobler and Wilson 1990). The impending competition between species for limited resource pools may force some species out of their fundamental niche space with inevitable local extinction.

Although resource niche width is an important concept in ecology, it is difficult to measure. Foraging, diet observations, and even genetic analysis of gut or faecal material are time consuming and often not feasible for some taxonomic groups. Further, these measures only provide a snapshot of the diet width of the organism's last meal. Stable isotopes analysis is often applied to indirectly measure the trophic resource use of species because the ratio of heavy to light nitrogen (N) increases with trophic position (DeNiro and Epstein 1981, Minagawa and Wada 1984). That is because ^{15}N is assimilated into consumer tissue at a higher rate than ^{14}N. Thus, the consumer has an elevated ratio of ^{15}N to ^{14}N ($\delta^{15}N$) compared to food items consumed and organisms at higher trophic levels have greater $\delta^{15}N$ than organisms at lower trophic levels (DeNiro and Epstein 1981, Minagawa and Wada 1984). $\delta^{15}N$ is also useful because it reflects a long-term picture of N that was incorporated into consumer tissues and not simply the consumer's last meal (Boecklen et al. 2011, Layman et al. 2012). The trophic niche width of a population is reflected in the variance around the mean of $\delta^{15}N$ (Bearhop et al. 2004), where greater variance in $\delta^{15}N$ reflects a population consuming a greater diversity of trophic resources

(Bearhop et al. 2004). Repeated measurements of individuals can capture temporal fluctuations in trophic position and temporal trophic niche width. Further, differences between mean $\delta^{15}N$ across habitats reflects a shift in trophic position (Nakagawa et al. 2007, Gibb and Cunningham 2011). For example, a shift in the diet of omnivorous ants can be observed as they switch between feeding as predators on other arthropods to feeding as herbivores through the consumption of plant nectar and honeydew resources produced by honeydew-producing hemipterans (Davidson et al. 2003, Tillberg et al. 2007, Blüthgen and Feldhaar 2010, Menke et al. 2010, Gibb and Cunningham 2011). Although much research has used stable isotopes to quantify trophic niche width, no study has applied this technique to capture potential changes in the trophic niche width of a species across an agricultural intensification gradient.

In this study, we compare the trophic niche width of four ant species across intensified (low shade) and non-intensified (high shade) coffee agroecosystems. Coffee is traditionally grown under a canopy of shade trees, however intensification reduces shade tree number, tree diversity, and canopy cover (Perfecto et al. 1996, Moguel and Toledo 1999). Intensification of coffee by eliminating shade trees drives the loss of many taxonomic groups, including ants, butterflies, birds and bats (Perfecto et al. 2007, Philpott et al. 2008). We investigate if coffee shade intensification alters the trophic position, trophic niche width, and the diet overlap between four omnivorous ant species. Ants are ideal study organisms because they feed from multiple trophic levels and competition is considered a hallmark of ant ecology (Hölldobler and Wilson 1990), thus shared resources are of utmost importance (Hölldobler and Wilson 1990). We predicted that trophic niche width (variance in $\delta^{15}N$) would decline in intensified plantations because of reduced resource diversity. We also predicted that ant species would shift trophic position to have more predatory signatures (higher $\delta^{15}N$) and that overlap in trophic niche width between species would increase in intensified, low-shade systems. We then provide analysis of one ant species, *Azteca sericeasur* Longino, to investigate the site level characteristics that drive the trophic resource use of this species and the influence of season on its temporal trophic niche width.

Methods

This study was conducted across 23 sites within six large coffee plantations varying in shade management in the Soconusco region of Chiapas, Mexico. Coffee plantations in the region are intensified by reducing shade tree density, diversity, and canopy complexity. We binned coffee plantations into two types: High shade plantations had >60% canopy cover and low shade had <35% canopy cover. In May-July of 2011, within each site, we searched the ground, coffee, and shade trees for four ant species: *A. sericeasur*, *Procryptocerus hylaeus* Kempf, *Pseudomyrmex simplex* Smith, and *Solenopsis geminata* Fabricius. *Azteca sericeasur* was previously referred to as *A. sericeasur*, but has been re-identified as *A. sericeasur* due in part to the queens' smaller ocelli and distinct yellow and brown facial markings (J. Longino, *pers. comm.*). *Solenopsis geminata* is a ground nesting species, *P. simplex* and *P. hylaeus* nest in hollow twigs in coffee and in shade trees, and *A. sericeasur* nests in shade trees. These species were selected to broadly represent the ant community and because they all are found in both intensified and non-intensified plantations. All species can be observed foraging in any of the three strata (ground, coffee, and shade trees). After hand collection, we stored ant workers in 96% alcohol before removing their gasters to avoid contamination of stable isotope analysis with gut contents. Alcohol storage did not influence δ^{15}N values (mean difference ± SEM, -0.0083±0.066, t = -0.126, df = 3, P = 0.908) (Blüthgen et al. 2003). We also collected grasshoppers and scale insects (*Octolecanium* sp. Kondo) to represent herbivores and we collected spiders to represent known predators in systems. To represent primary producers, we collected coffee leaf tissue at each site. We dried all samples at 60°C for 72h. To prepare samples for stable isotope analysis, we weighed approximately 725µg of animal tissues and 3622µg of plant tissues into tin capsules. Preliminary analysis found these weights to be optimal. Some ant colonies sampled had very few individuals; therefore we weighed whole ant bodies (without gasters) into capsules to avoid loss of material during transfer of ground material. The use of whole body (minus gaster) samples did not differ from ground ant samples without gasters (mean difference ± SEM, 0.34±0.17, df = 3, paired-t = -2.0, *P*=0.138).

To calculate δ^{15}N to estimate trophic niche position and width, we performed stable isotope analysis at the University of Michigan Stable Isotope Laboratory. Sample weight percent and isotopic analysis were performed using a Costech ECS 4010 elemental analyzer attached to a Finnigan Delta V+ mass spectrometer. Results were calibrated on a per-run basis using

international standards (IAEA600, USGS 25, IAEA N2). $\delta^{15}N/^{14}N$ are given in delta notation relative to atmospheric N, and precision of $\delta^{15}N$ was no worse than +/- 0.12 per mil. We calculated δ N as: [(sample $^{15}N/^{14}N$)/(standard $^{15}N/^{14}N$)-1] × 1000. Geographic variation influences $\delta^{15}N$, therefore we corrected animal samples by coffee samples because plant $\delta^{15}N$ reflects variation in geographic $\delta^{15}N$ (Gibb and Cunningham 2011); the correction was as follows: $\delta^{15}N_{corrected} = \delta^{15}N_{animal} - \delta^{15}N_{plant}$ at each site (ant versus plant $R^2 = 0.336$, $F_{1,22} = 10.6$, $P = 0.004$). It should be noted that all results are consistent regardless of the inclusion or exclusion of the correction factor.

To determine the differences in trophic resource use between species, we compared the mean and variance between ant $\delta^{15}N_{corrected}$ values in high and low shade plantations. We compared the trophic niche width by calculating the coefficient of variation (CV) for each species in high and low shade and compared the influence of intensification with a paired-test t. To compare shifts in trophic position and changes in the overlap in trophic niche width between species, we compared mean $\delta^{15}N_{corrected}$ across habitat type and ant species using a general linear mixed model (GLMM). We incorporated ant species, shade management, and the interaction between effects as fixed effects in the model. We incorporated site as a random effect in the model to account for pseudoreplication within site. To determine if the overlap in trophic resource use between species differed between shade management types, we analyzed pair-wise comparisons between species in high and low shade management using Bonferroni corrected t-tests of estimated marginal means (SPSS 21.0). Overlap between trophic resource-use between species was inferred by non-significant differences in mean $\delta^{15}N_{corrected}$ of ant species. We tested for normality by comparing histograms of data, the residuals of the model with q-q plots, and with Kolmogorov-Smirnov Tests. The CV was log transformed to improve the assumptions of normality.

To help explain trends in ant trophic position, we quantified five nest level factors for *A. sericeasur* ants for the rainy season 2011. We focused on *A. sericeasur* because it was the most sedentary-nested species. *(1) Shade tree honeydew-producing hemipterans* – For each nest tree, we cut 5 branches from at least four sides of the canopy. In all trees, sessile scale insects (Coccidae), mealybugs (Pseudococcidae), and whiteflies (Aleyrodidae) were the most dominant

honeydew-producing hemipterans. We standardized the number of honeydew-producing hemipterans recorded per branch by the estimated total leaf area of the branch sampled. *(2) Coffee honeydew-producing hemipterans* – We also used a modification of a scale insect survey, developed in Vandermeer and Perfecto (2006), to estimate the mean number of coffee green scales (*Coccus viridis*) per coffee bush on the nearest three coffee bushes to the nest tree. *(3) Nest tree type* – Shade tree species in the genus *Inga* produce extra-floral nectaries, which are tended by *A. sericeasur* and other ants and *A. sericeasur* also tends more honeydew-producing hemipterans when nesting in *Inga* spp. trees than in non-*Inga* spp. trees (data not shown). The *Inga* tree species included were, *I. micheliana*, *I. rodrigueziana,* and *I. vera*. The non-*Inga* tree species included were *Miconia affinis* (Melastomataceae), *Alchornea latifolia* (Euphorbiaceae), *Trema micrantha* (Cannabaceae), *Yucca elephantipes* (Asparagaceae), *Syzygium jambos* (Myrtaceae)*, Cordia stellifera* (Boraginaceae)*,* and *Ocotea* sp. (Lauraceae). Therefore, we categorized nest trees as *Inga* or non-*Inga* species. *(4) Nest tree connections* – We counted the number of trees that came into contact with a branch or trunk of the nest tree (hereafter nest tree connections). Having more nest tree connections allows colonization of multiple trees. *(5) Nest activity strength* – As a proxy for nest size, we measured nest activity strength. First, we beat nest tree trunks and videotaped ant activity on three index cards (7.6×12.7cm) pinned to areas with high ant activity. We count the number of ants per card at time 10s and 20s of the video footage and averaged across all cards per nest (nest activity strength). We used GLMM to determine which of the five site variables explained $\delta^{15}N_{corrected}$ of *A. sericeasur*. We used Akiake Information Criteria (AIC) to determine the best-fit model and eliminated non-significant factors that reduced model fit. We treated site as a random effect in the model, however it did not improve model fit and was removed.

To test for any temporal effects on trophic niche width and any interactions between shade management and season, we focused on *A. sericeasur* because it was possible to resample nests over time. After initial collection from the rainy season of 2011 (N = 34), we sampled nests again in the dry (Mar ‘12) (N = 34) and rainy season (Jun-Jul ‘12) (N = 33). Because some nests disappeared, we incorporated 7 new nests in the rainy season of 2012. We measured $\delta^{15}N$ as described above. To test the hypothesis that shade management modifies the effect of season on *A. sericeasur* trophic position, we used repeated-measures GLMM to test effects. We

incorporated site as random effect in the model with nest as the repeated subject. We included season, shade management, and their interaction as fixed effects. We compared differences between season using pair-wise estimated marginal means (t-tests). In addition to this analysis, we also calculated the temporal niche width for all nests with 2 or 3 measurements (N = 23) by calculating the CV for each nest. We compared the mean CV in high and low shade sites with a general linear model. We confirmed normality as described above. All analyses were conducted in SPSS (21.0).

Results

The trophic niche width of all species was reduced with intensification; the mean CV $\delta^{15}N_{corrected}$ was smaller in low shade relative to high shade coffee plantations (Fig. 3.1; mean difference ± SE, 0.37±0.15, df = 3, paired-t = 3.56, P=0.038). Further, the overlap in trophic niche width was modified by intensification. There was a significant ant species by shade management interaction such that the $\delta^{15}N_{corrected}$ of the four ant species differed in response to shade intensification (Table 3.1, Fig. 3.1). Pair-wise comparisons revealed that under high shade management, *A. sericeasur* $\delta^{15}N_{corrected}$ was significantly lower than *S. geminata,* however under low shade conditions there was no difference between the means of the two species (Table 3.2). Conversely, *A. sericeasur* $\delta^{15}N_{corrected}$ was significantly higher than *P. simplex* in low shade management, however under high shade conditions there was no difference between the means of the two species (Table 3.2). This result suggests that pair-wise competitors for trophic resources may be modified by intensification. All other pair-wise comparisons between species were significantly different (Table 3.2). Pair-wise comparison also revealed that *A. sericeasur* $\delta^{15}N_{corrected}$ was 28% higher in low shade relative to high shade sites, but this difference was only marginally significant (mean difference ± SE: -1.1±0.5, P = 0.053). The trophic position of all other species did not significantly differ between high and low shade management, but $\delta^{15}N_{corrected}$ of *S. geminata* and *P. simplex* tended to decline with intensification. Comparing ant $\delta^{15}N_{corrected}$ to organisms at known trophic levels suggested that *S. geminata, A. sericeasur,* and *P. simplex* had trophic positions near to predatory spiders, while *P. hylaeus* had a position near to known herbivores (Fig. 3.1).

Site level analysis of *A. sericeasur* nests determined that the number of tree connections to the nest tree best explained the $\delta^{15}N_{corrected}$ of *A. sericeasur* (Table 3.3, Fig. 3.2A). Surprisingly, shade tree type, the density of honeydew-producing organisms in shade tree canopies and in coffee, and nest activity strength did not explain significant variation in *A. sericeasur* $\delta^{15}N_{corrected}$ (Table 3.3). Thus, it appears the use of multiple trees is important to trophic resource use. Importantly, nest trees in high shade plantations had more arboreal connections than nest trees in low shade plantations (Fig. 3.2B, $df_{num,den}$=1,14, F = 15.4, *P*=0.002).

Comparing the influence of shade management across multiple seasons revealed a significant shift of *A. sericeasur* in trophic position and across season (Table 3.4, Fig. 3.3). *Azteca sericeasur* nests in low shade plantations had 33% higher $\delta^{15}N_{corrected}$ than did nests in high shade plantations. Pair-wise comparison of estimated means revealed *A. sericeasur* in the rainy season '12 had $\delta^{15}N_{corrected}$ values 10% lower than in the rainy season '11 (mean difference ± SE: -0.38±0.1, P < 0.001), and 15% lower in the dry season '12 (mean difference = -0.56±0.1, P < 0.001). The rainy season '11 was marginally lower than the dry season '12 (mean difference = -1.03±0.54, P = 0.066). *Azteca sericeasur* temporal trophic niche width did not differ between nests in high (mean CV $\delta^{15}N$ ± SE: 0.101 ± 0.021) and low shade (mean CV $\delta^{15}N$ ± SE: 0.096 ± 0.009) management ($F_{1,30}$ = 0.07, P = 0.797).

Discussion

This study is the first to show that agricultural intensification reduces trophic niche width, alters the niche overlap between species, and is among the first to show that intensification shifts trophic position. Agricultural intensification is a strong anthropogenic driver of biodiversity loss (Tilman et al. 2001, Bengtsson et al. 2005, Tscharntke et al. 2005, Perfecto et al. 2009) and changes to ecosystem processes (Matson et al. 1997). The goal of intensification is often to simplify the physical and biological components of agro-ecosystems to make way for maximum production of a sole crop species. This simplification limits resource pools for species at low and high trophic levels. Species feeding directly or indirectly as herbivores are restricted to feeding from fewer plant species, and likewise, for predators prey abundance and diversity often decline in intensified systems (Wickramasinghe et al. 2004, Morris et al. 2005, Taylor 2008, Mandelik et al. 2012, Frey-Ehrenbold et al. 2013). Previous work demonstrates that other environmental and

anthropogenic factors alter trophic niche ecology of species. Land-use change impacts ant trophic position (Gibb and Cunningham 2011), logging of primary forest limits the trophic niche width and position of birds (Edwards et al. 2013), fragmentation of tidal creeks reduces the niche width of a top predatory fish (Layman et al. 2007), and habitat characteristics of islands alter the trophic niche width of rats (Rodriguez and Herrera 2013). Like these other factors, intensification impacts ant community structure through the alteration of trophic resource use, however the mechanisms driving these changes are not always clear.

The mechanism behind the collapse of ant trophic niche width in intensified coffee may be complex and possibly different for each species. The reduction of shade tree number, canopy size, and connectedness limits the three-dimensional size of the coffee agroecosystem. Further, shade trees provide a number of structural features that make coffee plantations forest-like, which also likely impact prey and mutualist populations. Ecosystem size and structural floral features are hypothesized to help explain the effects of habitat fragmentation on predatory fish niche width (Layman et al. 2007). Ant trophic position and nest site location may cause different species to capture different trophic resources (Kaspari and Yanoviak 2001). *Solenopsis geminata, A. sericeasur,* and *P. simplex* have δ^{15}N signatures that are comparable to known predators, while *P. hylaeus* is similar to known herbivores. Further, *S. geminata* is a ground-nesting species, whereas *P. hylaeus, P. simplex,* and *A. sericeasur* are arboreal nesting species. Honeydew and extra-floral nectar are resources that may be less available to species in low shade habitats. *Solenopsis geminata* also consumes seeds, therefore variability in this plant resource likely impacts niche width (Carroll and Risch 1984). At the same time, reduce trophic niche width of an omnivore can also occur if prey resources, like prey that are predators, become less available. However, it is likely that the combined loss of high and low trophic resources narrow the niche widths of omnivorous ant species and it is difficult to speculate which specific resources are important to these changes.

This study is also among the first to show that agricultural intensification drives trophic shifts. Across three sampling seasons, we show that *A. sericeasur* trophic position is lower in high shade relative to low shade plantations. Further, site level analysis also suggests that greater nest-tree canopy connectivity drives lower trophic positions. One could argue that the loss of canopy

access reduces the availability of basal trophic resources (honeydew) for *A. sericeasur*. Indeed, Gibb and Cunningham (2011) support this hypothesis in a study of different land-use types. They show that land-use change from pasture to re-vegetated pastures with young trees, lowers the community wide trophic position of ant genera (Gibb and Cunningham 2011). However, we did not observe a community wide decline in trophic position. *Solenopsis geminata* and *P. simplex* tended to have lower trophic positions in low relative to high shade plantations, the reverse pattern of *A. sericeasur*. Therefore it appears that more vegetation does not always drive species to reside at lower trophic levels.

The opposing directions of trending ant trophic shifts across management types resulted in a change in trophic niche overlap between species. We argue that overlaps in trophic niche width are especially important for communities that are structured by competition for resources. The distribution of ant species is often influenced by interspecific competition (Hölldobler and Wilson 1990, Parr and Gibb 2010) and much research at the study site documents competition for food, nesting sites, and spatial resources (Ennis 2010, Philpott 2010, Perfecto and Vandermeer 2013). We show that *A. sericeasur* δ^{15}N overlaps with *S. geminata* in low shade habitats, but overlaps with *P. simplex* in high shade habitats. For *A. sericeasur* in low shade habitats, the lack of foraging area in the nest tree canopies may drive workers to forage on the ground, where they may have greater overlap in resource use with *S. geminata*. In accordance, previous research suggests that in low shade plantations *A. sericeasur* negatively impacts ground-foraging ant diversity to a greater extent than in high shade plantations where *A. sericeasur* promotes local ant diversity (Ennis 2010). Together our results paint a complex picture, where the loss of shade tree canopies drives *A. sericeasur* to forage in lower strata and increase overlap of trophic resource use with ground-nesting species, which coincides with the displacement of some ground-foraging ants (Ennis 2010).

The intensification of agriculture is one the most important environmental issues of the modern era largely because it drives the loss of species and disrupts ecosystem processes (Matson et al. 1997, Tilman et al. 2001, Bengtsson et al. 2005, Attwood et al. 2008). Many organisms, including ants, living in agricultural landscapes provide important services to agriculture including: soil improvement, pollination, and pest control (Daily 1997, Wielgoss et al. 2014).

Azteca sericeasur, P. simplex, P. hylaeus, and other ant species consume and remove important pests from coffee (Perfecto and Vandermeer 2006, Philpott et al. 2012, Gonthier et al. 2013). Changes to ant trophic ecology may have important consequences for the quality of the pest control services these ants provide, however more work is needed to describe these connections. This study is one of the first to elucidate many changes to ant trophic ecology following intensification. It is the first to show that agricultural intensification reduces trophic niche width, alters the niche overlap between species, and is among the first to show that intensification can result in shifts in trophic position. These findings help describe the ecological changes that accompany intensification and will be useful in uncovering the mechanism behind the loss of biodiversity.

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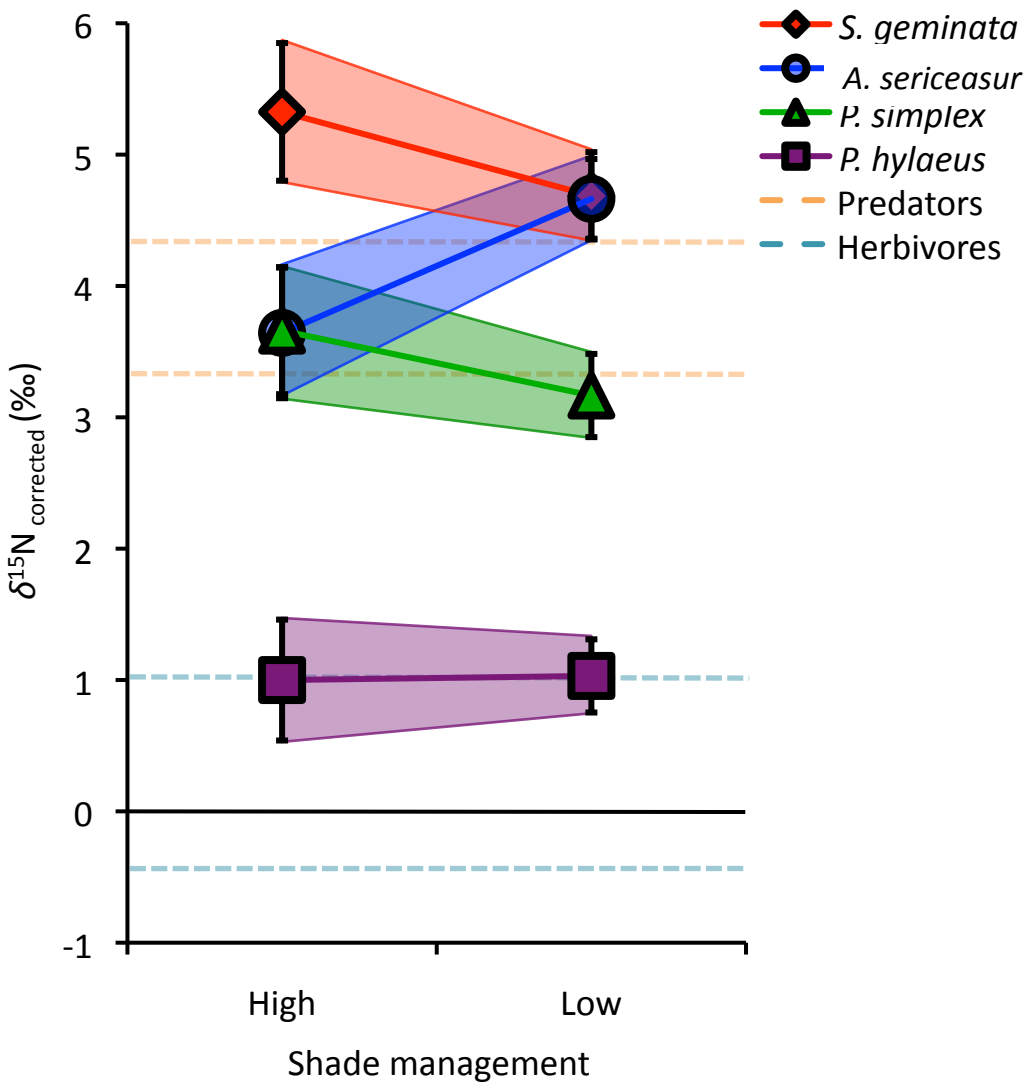

Figure 3.1. Mean (± SE) trophic niche position ($\delta^{15}N_{corrected}$ [‰]) of ant species under high and low shade management. Orange lines indicate known predator (spiders), and turquoise lines indicate known herbivore (grasshoppers and scale insects) upper and lower SE limits of their $\delta^{15}N_{corrected}$ distributions. $\delta^{15}N$ values were corrected by subtracting coffee plant $\delta^{15}N$ at each site.

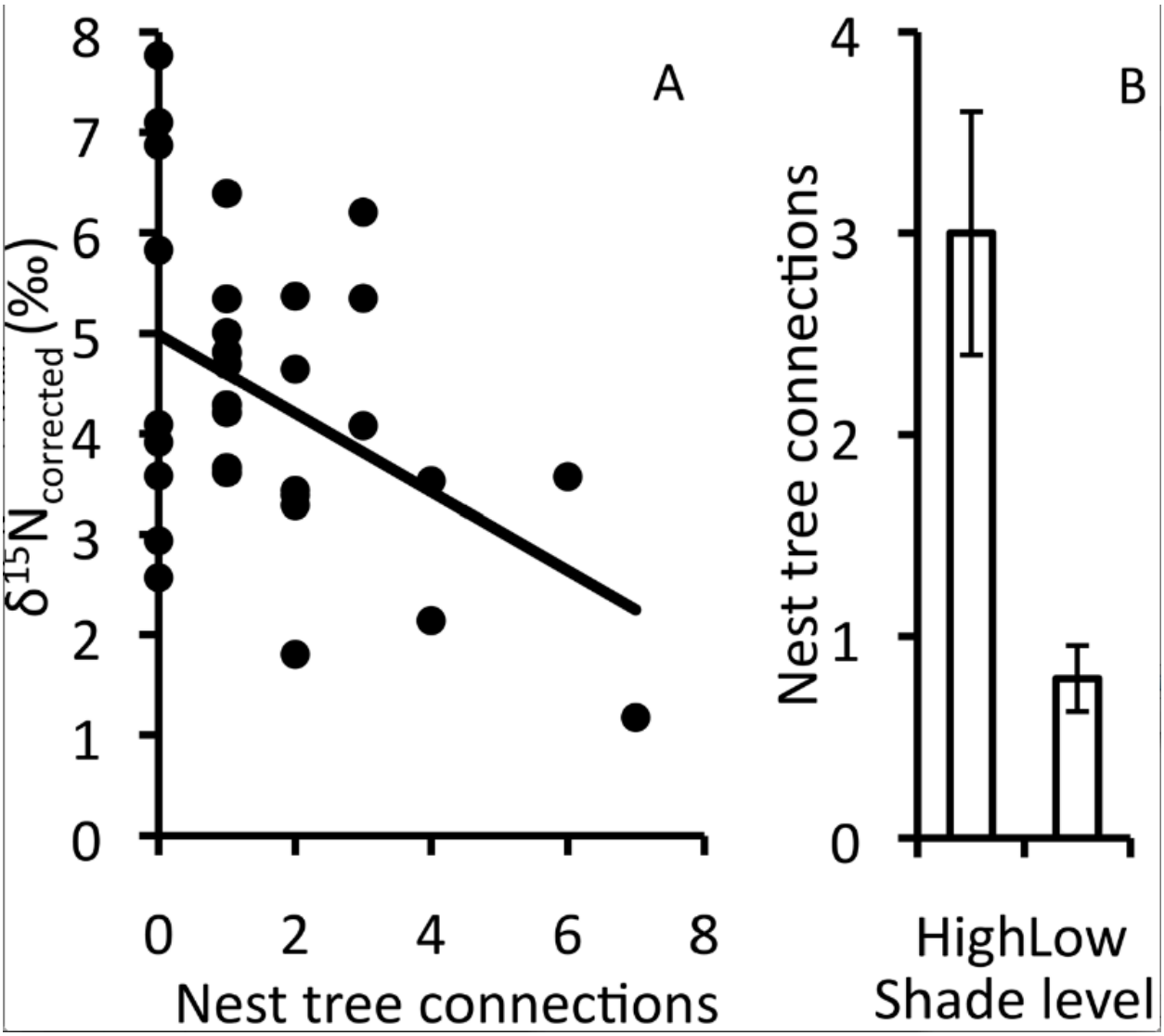

Figure 3.2. (A) Relationship between the number of arboreal connections between *A. sericeasur* nest tree and neighbouring trees and *A. sericeasur* trophic position ($\delta^{15}N_{corrected}$ [‰]). (B) Mean (± SE) number of arboreal connections on *A. sericeasur* nest trees in high and low shade plantations. Ant $\delta^{15}N$ was corrected by subtracting coffee plant $\delta^{15}N$ at each site.

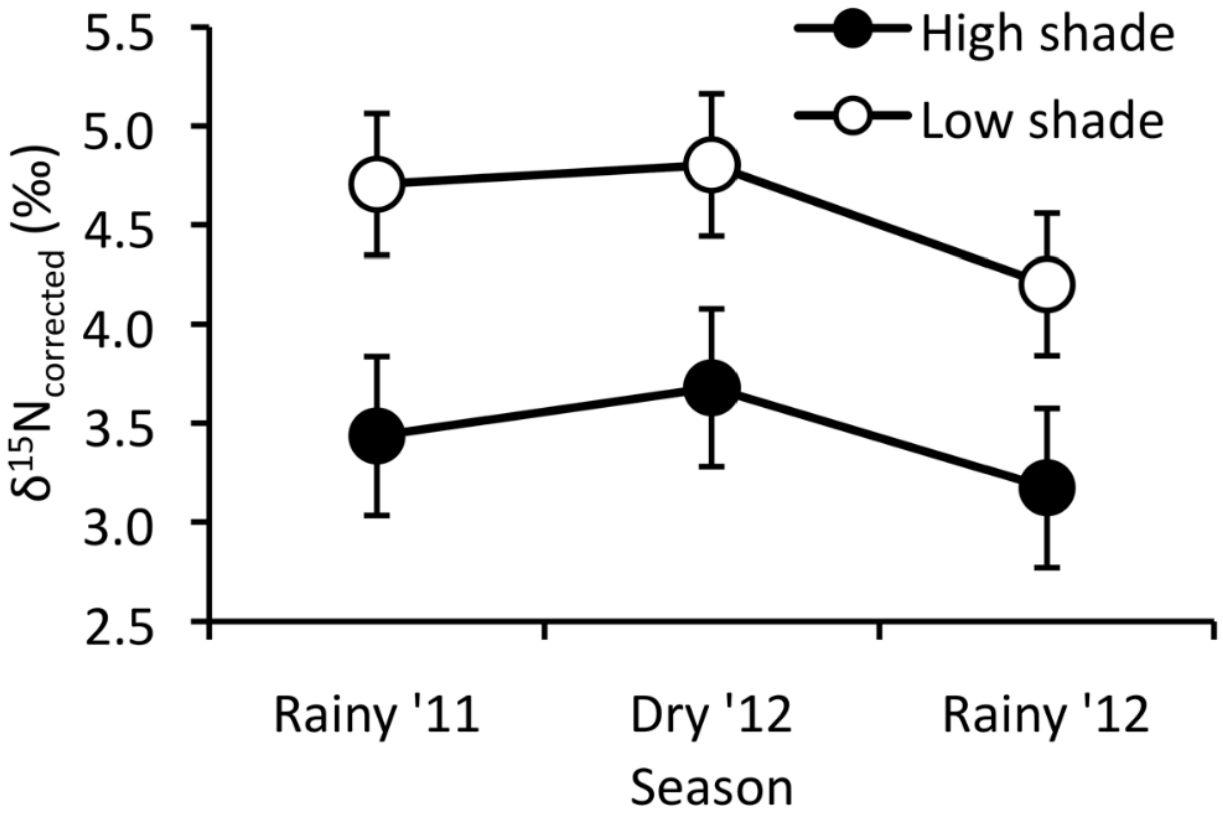

Figure 3.3. Estimated mean (±SE) trophic position ($\delta^{15}N_{corrected}$ [‰]) in high and low shade management across season for *A. sericeasur*. Ant $\delta^{15}N$ was corrected by subtracting coffee plant $\delta^{15}N$ at each site.

Table 3.1. Ant species and shade management effects on ant $\delta^{15}N_{corrected}$ (‰).

	df*	F	P
Intercept	1,23	218.8	<0.001
Shade	1,23	0.1	0.763
Ant	3,73	59.7	<0.001
Shade × Ant	3,73	3.8	0.014

*df = Numerator df, Denominator df

Table 3.2. Mean difference ± 95% confidence limit in estimated marginal means for pair-wise comparisons of species $\delta^{15}N_{corrected}$ in high and low shade management.

High shade	*P. hylaeus*	*P. simplex*	*S. geminata*
A. sericeasur	**3.078±1.306***	-0.031±1.206NS	**-1.704±1.147***
P. hylaeus		**-3.109±1.383***	**-4.782±1.352***
P. simplex			**-1.673±1.235***
Low shade	*P. hylaeus*	*P. simplex*	*S. geminata*
A. sericeasur	**3.51±0.984***	**1.62±1.002***	-0.156±1.15NS
P. hylaeus		**-1.891±1.145***	**-3.666±1.234***
P. simplex			**-1.776±1.249***

*Bold = significant difference between means; non-bold = non-significant difference between means.

Table 3.3. Site level factors explaining mean *A. sericeasur* $\delta^{15}N_{corrected}$ (‰) for 2011 rainy season.

	df*	F	P
Intercept	1,27	137.8	<0.001
Nest tree connections	1,27	7.8	0.009
Tree type	1,27	2.0	0.172
Shade tree honeydew-producing hemipterans	2,27	0.7	0.418
Coffee honeydew-producing hemipterans	-	-	-
Nest activity strength	-	-	-

*df = Numerator df, Denominator df. Dashes indicate factor was non-significant and was removed from the best-fit model by comparison of AIC.

Table 3.4. Season and shade management effects on mean *A. sericeasur* $\delta^{15}N_{corrected}$ (‰).

	df*	F	P
Intercept	1,27	230.7	<0.001
Season	2,58	18.0	<0.001
Shade	1,27	4.7	0.039
Season × Shade	2,58	0.8	0.454

*df = Numerator df, Denominator df.

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Chapter 4
Ants defend coffee from berry borer colonization

Abstract

Ants frequently prevent herbivores from damaging plants. In agroecosystems they may provide pest suppression services, although their contributions are not always appreciated. Here we compared the ability of 8 ant species to prevent the coffee berry borer from colonizing coffee berries with a field exclusion experiment. We removed ants from one branch (exclusion) and left ants to forage on a second branch (control) before releasing 20 berry borers on each branch. After 24 hours, 6 of 8 species had significantly reduced the number of berries bored by the berry borer compared to exclusion treatment branches. While the number of berries per branch was a significant covariate explaining the number of berries bored, ant activity (that varied greatly among species) was not a significant factor in models. This study is the first field experiment to provide evidence that a diverse group of ant species suppress the berry borer from colonizing coffee berries.

Introduction

Some species of ants benefit plants (Styrsky and Eubanks 2007, Chamberlain and Holland 2009, Rosumek et al. 2009, Trager et al. 2010). Humans have known this for quite a long time. In fact, ants were described as biological control agents in China around 304 AD (Van Mele 2008). Many plants have also evolved to promote the activity of ants on their tissues. Surveys of tropical forests show that up to one third of all woody plants have evolved ant-attracting rewards (Schupp and Feener 1991). Some plants provide domatia as ant housing structures, while others attract ants to their tissues with extra-floral nectaries. Some plants are hosts to honeydew-producing hemipterans that excrete honeydew, a sugary substance consumed by ants. Still other plants are simply substrates for ant foraging. The majority of studies conducted across these ant-plant groups show that ants benefit plants by removal of herbivores (Chamberlain and Holland 2009, Rosumek et al. 2009, Trager et al. 2010). Nonetheless, in many agroecosystems, the

benefits of pest control services by ants are not recognized. Agricultural managers often view them as pests or annoyances to agricultural production because some ants tend honeydew-producing insects that can damage crops (Styrsky and Eubanks 2007, 2010). However, a review of the literature on ant-hemipteran associations suggests that even these associations benefit plants indirectly because ants remove other, more damaging herbivores (Styrsky and Eubanks 2007, 2010). Regardless, the literature lacks studies investigating ant-plant interactions in agroecosystems. Here, we broadly survey the pest suppression services provided by a suite of ant species to better understand the role of ant defense of coffee.

Coffee is a tropical crop that occurs as an understory shrub in its native range, and coffee plants are therefore often grown under a canopy of shade trees in agroforesty systems in some parts of the world (Perfecto et al. 1996). This canopy layer provides plantations with a forest-like vegetation structure that can help maintain biodiversity (Perfecto et al. 1996). Ant biodiversity is high in many coffee plantations and ants attack and prey on many coffee pests, including the coffee berry borer (CBB; *Hypothenemus hampei* [Ferrari] [Coleoptera: Scolytidae]) (Philpott and Armbrecht 2006, Armbrecht and Gallego 2007, Vandermeer et al. 2010). For example, *Azteca sericeasur* Longino (previously referred to as *A. instabilis*) is a competitively dominant ant that aggressively patrols arboreal territories in high densities and previous research has found that it impacts the CBB (Perfecto and Vandermeer 2006, Larsen and Philpott 2010). Some laboratory and observational field studies have found that *Pseudomyrmex* spp., *Procryptocerus hylaeus* Kempf*,* and *Pheidole* spp. may suppress the CBB (Jiménez-Soto et al. 2013, Larsen and Philpott 2010, Philpott et al. 2012). However, other field experiments have not found ants to be biological control agents of the CBB (Varón et al. 2004, Vega et al. 2009). Further, the pest control effects of many ant species on the CBB have not yet been evaluated and it could be that previously documented effects are specific to only a few species.

Chemical insecticides used to control CBB are not always effective (Vega et al. 2006) because the CBB lifecycle takes place largely hidden within coffee berries (Vega et al. 2006) and the CBB has developed insecticide resistance (Brun et al. 1990). For that reason, natural ant pest control of the CBB is particularly important. Several of the stages of the CBB life cycle make it vulnerable to attack by ants (Damon 2000, Jaramillo et al. 2007). First, the CBB hatches from

eggs within the coffee berry, where it consumes the seeds (Damon 2000, Jaramillo et al. 2007). Small ants may enter the berry through the beetle entrance hole and predate the larvae and adults inside (Larsen and Philpott 2010, Perfecto and Vandermeer 2013). Second, old berries infested with the CBB may not be harvested because they often turn black and remain on the coffee branches or may fall to the ground (Damon 2000, Jaramillo et al. 2007). These old infested berries may act as a reservoir of borer populations and ant predation at this stage could be very important for suppressing CBB populations in the next season. Third, as adult borers disperse (flying or crawling) to colonize new berries, ants may prevent them from entering new berries (Perfecto and Vandermeer 2006, Philpott et al. 2012). To date, no field experiment has specifically investigated how coffee-foraging ants suppress CBB colonization of berries. Here, we studied the abilities of eight ant species to prevent colonization of berries by the CBB. We hypothesized that only species with high activity on branches would suppress CBB colonization of berries. We show that 6 of 8 ant species suppress CBB colonization of berries and that the effect of ants is independent of ant activity on branches. This study is the first field experiment to provide evidence that a diverse group of ant species suppress the CBB from colonizing coffee berries.

Methods

Our research was conducted on Finca Irlanda, a coffee plantation in the Soconucsco region of southern Mexico and the site of much ongoing research regarding community ecology of the arthropod interaction web (Vandermeer et al. 2010). In this region, the CBB is a major pest of coffee (Vandermeer et al. 2010). We searched for coffee bushes occupied by one of eight species that were each abundant enough to obtain sufficient replication for this experiment: *A. sericeasur* ($N = 20$), *Crematogaster* spp. ($N = 20$), *Pheidole synanthropica* Longino ($N = 19$), *Pseudomyrmex simplex* Smith ($N = 30$), *Pseudomyrmex ejectus* Kempf ($N = 28$), *Solenopsis picea* Emery ($N = 31$), *Tapinoma* sp. ($N = 30$), and *Wasmannia auropunctata* Roger ($N = 28$) (N = sample size of bushes used in experiment). *Azteca sericeasur* was previously referred to as *A. sericeasur*, but has been re-identified as *A. sericeasur* due in part to the queens' smaller ocelli and distinct yellow and brown facial markings (J. Longino, *pers. comm.*). Our goal was to capture a broad survey of the ant species that occupy the coffee vegetation in the coffee plantation. Within the plantation, five *Crematogaster* spp. forage in the coffee, however field

identification at the time was not reliable therefore taxonomic resolution for *Crematogaster* spp. remained at the genus level. For *P. simplex* and *P. ejectus* it was not always possible to find occupied bushes by observation of ant foraging. Instead, for *P. simplex* and *P. ejectus*, we determined occupation by removing all dead twigs on the coffee bush and searching these for ant nests within the hollow branches (Philpott and Foster 2005). We reattached the nested hollow branch to a living branch with thin wire and treated these bushes as bushes occupied by *P. simplex* or *P. ejectus*.

To test the effects of each ant on CBB colonization of berries, we performed an ant exclusion experiment. We surveyed bushes occupied by one of the eight target ant species. We excluded coffee bushes with few branches to control for the size of the foraging area of each ant species. On each bush, we searched for two branches of equal age and position and roughly the same number of coffee berries (never more than 8 berries difference). On each branch, we removed all berries that had CBB entrance holes. We then removed all ants from one branch and applied tanglefoot (exclusion) to the base of the branch near the coffee trunk. On the second branch, we left ants to forage freely (control). To estimate ant activity, we counted the total number of ants foraging on the stem, leaves, and berries of each branch for 1-minute including those that travelled onto the branch during the 1-minute survey. We also counted ants on exclusion branches after the experiment and if a branch had more than one ant individual present, we excluded the bush from analysis (this occurred in only 2 cases). To release CBB onto control and treatment branches, we created a leaf platform to aid their chances of encountering berries. The leaf platform consisted of a coffee leaf that we cut in two places on one side of the leaf. The leaf was wedged between the branch stem and a cluster of berries to create a platform surrounding the cluster (Fig. 4.1a,b). A coffee leaf was used as a platform because artificial structures attract attention from many ant species. After waiting several minutes to ensure normal ant activity, we released 20 CBBs on the leaf platforms of the control and exclusion branches. After 24 hours, we counted the number of berries per branch that had CBBs inside entrance holes. We did not count partially bored holes in berries, nor CBBs that had bored into twigs and leaves. Multiple bored entrance holes per berry were only counted as one bored berry. We modified the experiment slightly for *P. simplex* and *P. ejectus* because of the difficulty in locating these species within a bush using visual cues (see above). For these two species, we used the living branch to which the

nest was attached as the control branch (with ants). This was done because we wanted to make sure that ants were actively foraging on control branches after the disturbance of removing nests.

To statistically analyze experimental data, we opted to use linear mixed models instead of paired t-tests because mixed models allow inclusions of experimental non-independencies through the incorporation of covariates. We included bush as a random effect in the model to pair control and exclusion branches within each bush. Ant species (each of the 8 ant species) and treatment (control or exclusion) and the species by treatment interaction were included as fixed effects in the model. To control for differences between each branch and bush, we included the number of berries per branch, the number of berries in contact with the leaf-platform, and the number of worker ants per branch (ant activity) as covariates in the model. We performed type III F-tests of significance for main effects with maximum likelihood (ML) to estimate the fixed effect parameters and variance of random effects (West et al. 2007). We removed non-significant factors from models and compared nested and null models with Likelihood Ratio Tests to determine the best-fit model. We also compared ant activity (per minute) across different species to determine if this factor might correlate with berries bored and vary across ant species. To determine if ant activity correlated with the number of coffee berries bored, we limited the dataset to only control branches (with ants) and used a generalized linear model with a Poisson log-link function because data did not meet the assumptions of normality. To determine if ant activity varied by species, we again limited the dataset to only control branches and used Analysis of Variance with Tukey's HSD analysis. We tested the normality of the data with qq-plots and Kolmogorov-Smirnov tests of model residuals. We conducted all statistical analyses with SPSS (20.0).

Results

The linear mixed model showed that the number of berries bored varied by ant species ($F_{7,206}$ = 3.5, P = 0.0013), exclusion treatment ($F_{1,208}$ = 44.9, P = 0.0001), and by the number of berries per branch ($F_{1,210}$ = 7.8, P = 0.0058). There was no interaction between ant species and treatment ($F_{7,206}$ = 1.8, P = 0.0961). Overall, pooling all ant species together, there were 50% more berries bored in exclusion branches relative to controls (Fig. 4.2). Pair-wise comparison of control (ant) and exclusion (no ant) branches revealed that six of 8 ant species significantly reduced the

number of berries bored relative to controls. On *A. sericeasur* control branches, there were 88% fewer CBB in berries, with *P. synanthropica* there were 200% fewer, with *P. ejectus* there were 66% fewer, with *P. simplex* there were 43% fewer, with *Tapinoma* sp. there were 210% fewer, and with *W. auropunctata* there were 86% fewer bored berries relative to their paired exclusion branches (Fig. 4.2). There was no difference between the number of bored berries on control and exclusion branches on bushes with *Crematogaster* spp. and *S. picea.* The number of berries in contact with the leaf platform and the number of ants per branch had no correlation with the number of berries bored.

Ant activity (ants/branch/minute) did not correlate with the number of berries bored (Wald χ = 1.6, df = 1, P = 0.204), but did differ across species when only control branches were considered ($F_{7,206}$ = 25.6, P = <0.0001). Across species, *Tapinoma* sp. and *W. auropunctata* had the highest activity, *A. sericeasur, Crematogaster* spp., *P. synanthropica,* and *S. picea* had intermediate activity, and *P. ejectus* and *P. simplex* had the lowest activity (as determined by Tukey's HSD; Fig. 4.3).

Discussion

Our study represents one of the first field experiments showing that a broad survey of ants reduce colonization of coffee berries by the CBB. This is in contrast to previous studies that suggest ants may not have any effects on CBB, especially in field experiments (Varón et al. 2004, Vega et al. 2009). Our results are in accordance with other observational studies that show that specific ant species may suppress CBB in coffee plantations, yet these studies have either focused on the most dominant or abundant species observed (Jiménez-Soto et al. 2013, Perfecto and Vandermeer 2006, Perfecto and Vandermeer 2013) or investigated the broad community-wide impacts of ants on the CBB (Larsen and Philpott 2010). Our experimental approach is limited to our understanding of how ants suppress CBB colonization of berries and not other life stages of the CBB. Our study suggests that ant occupation of coffee bushes is very important during a seasonal period when new coffee berries develop and the CBB begins to disperse from old infested berries to developing un-infested berries (Damon 2000).

It is surprising that *Crematogaster* spp. and *S. picea* did not suppress the colonization of berries, considering that other studies have shown species within these two genera have important effects on herbivores (Kaplan and Eubanks 2005, Philpott et al. 2008). Low ant activity on coffee bushes with *Crematogaster* spp. or *S. picea* cannot explain these results because these species had greater activity per branch than *P. ejectus* and *P. simplex* and equivalent activity to *A. sericeasur* and *P. synanthropica*, species that did suppress CBB damage. One explanation could be that because we grouped five *Crematogaster* spp. together into a single treatment, effects of individual species may be masked. For *S. picea,* it may be that under conditions of higher ant activity, with closer proximity to nest entrances, this species has effects, but this species does not have strong effect at lower activity. This species also has a small body size and moves relatively slowly in comparison to the species that did have an effect, which might have limited it from removing or easily capturing CBBs. *Wasmannia auropunctata* is of similar size to *S. picea* and still had strong effects on CBB. However, *W. auropunctata* had significantly higher ant activity levels on the branch as compared to *S. picea*. Perhaps the combination of low activity, small body size, and slower movement limited *S. picea* from affecting the CBB. While we found no effect of *S. picea* on CBB colonization of berries, it may be that *S. picea*, and other smaller ants, have important impacts on the CBB at other stages of the CBB life cycle because they can pass into entrance holes of CBB (Perfecto and Vandermeer 2013).

Experiments with both *P. simplex* and *P. ejectus* employed slightly different methodologies than the other ant species, which may have intensified the effect of these ants. For these two species, hollow twigs that contained ants were attached to a branch with berries and this branch was used as the control branch in the experiment. This likely elevated the number of ants/branch/minute. However, in the lab, *P. simplex* had similar effects on the CBB (Philpott et al. 2012). Additionally these two species had the lowest densities on control branches of all other species; averaging 3.6 and 3.7 ants per branch for *P. ejectus* and *P. simplex,* respectively. Thus, these species have effects at very low numbers, and the results of this study should only pertain to branches for which the density of these species reaches this mark.

Certain aggressive ants (that spatially defend arboreal territories) that suppress CBB colonization of berries might also benefit CBB after colonization. Larger ants cannot enter berries, but if they

are aggressive competitors for space, they will prevent other ants from occupying the branches they patrol (Perfecto and Vandermeer 2013). These ants, likely *A. sericeasur* and *P. synanthropica*, may provide CBB with enemy free space after the CBBs colonize berries in their territories.

In conclusion, we find that 6 of 8 ant species suppressed CBB colonization of coffee berries suggesting that ants generally provide important pest suppression services within coffee agroecosystems. This is the first field experiment to demonstrate general ant suppress CBB colonization. This finding is important considering that chemical pesticides are thought to be ineffective at controlling the CBB (Brun et al. 1990, Vega et al. 2006). Nonetheless, ants do not completely control the CBB, other control agents like birds, parasitoids, and fungal pathogens also aid in the control of the CBB (Vega et al. 2009). Further work should look at larger scale impacts of ants on the CBB, such as farm scale impacts. Also, more theoretical work is needed to understand how ants impact the CBB at different stages of its life cycle and to reveal which stage of the life cycle is most important for population regulation. Nonetheless, this study provides strong evidence that ants defend coffee from CBB colonization.

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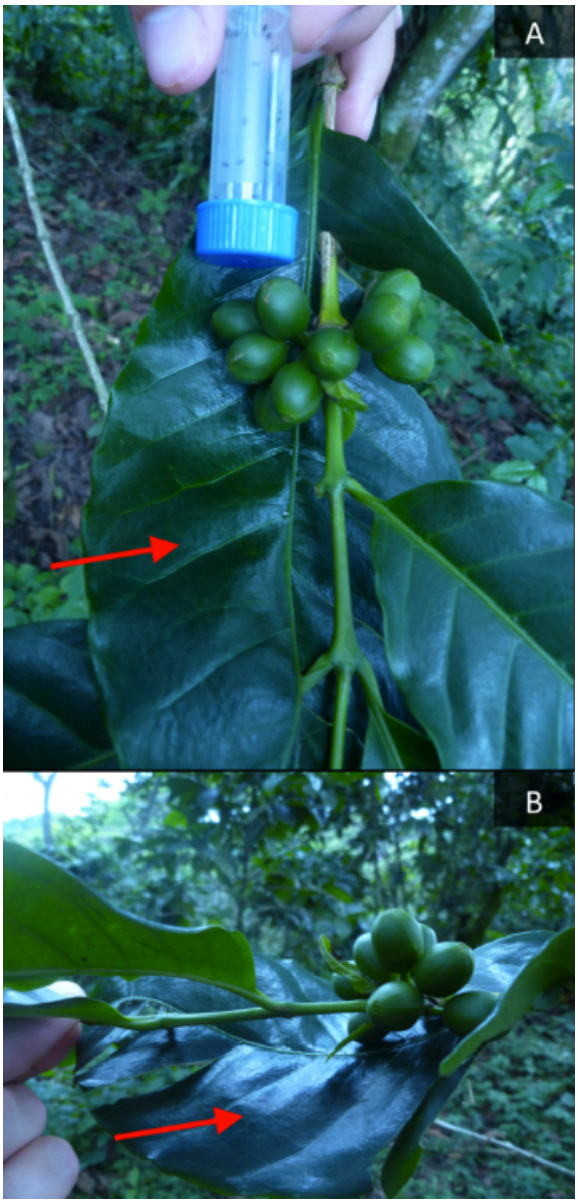

Figure 4.1. Photographs of the leaf platform attached for the release of the CBB on control and treatment branches. a) View from above with a vial of CBBs. b) Horizontal view. Arrows point to the leaf platform.

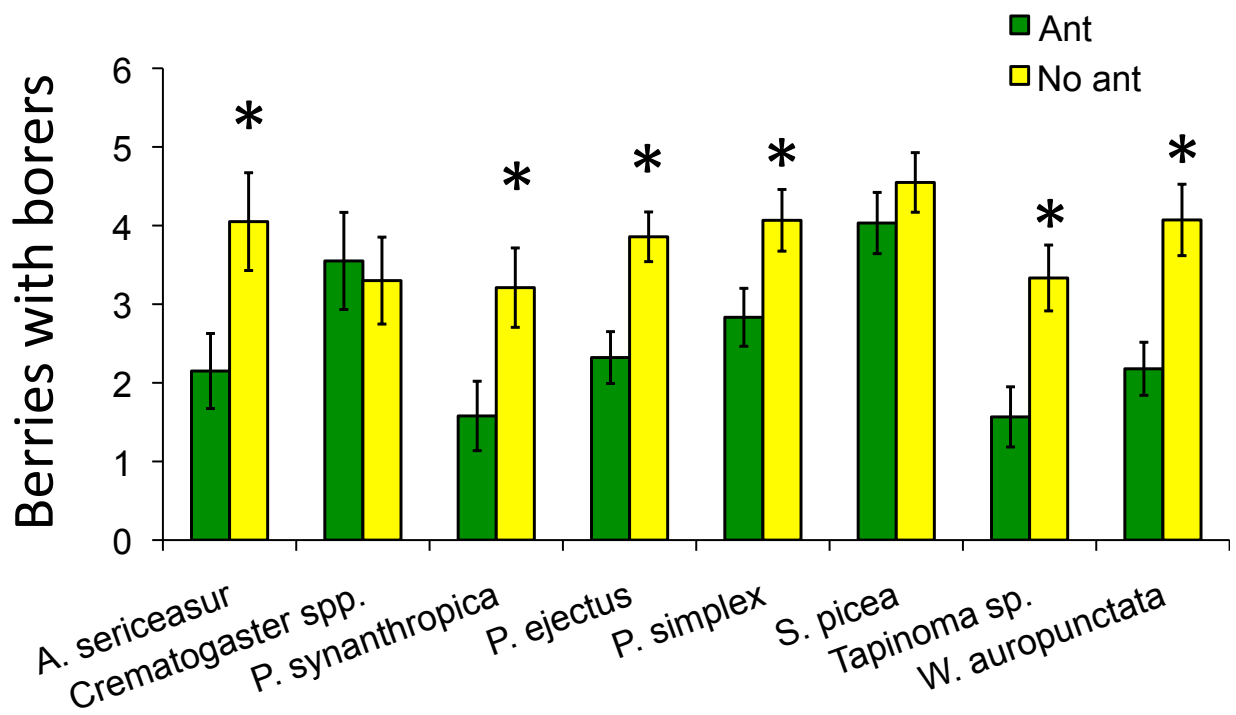

Figure 4.2. Means and standard errors of the number of berries bored by CBBs (per branch) across ant species treatments and ant-exclusion treatments. Asterisks indicate significant differences between control (ant) and exclusion (no ant) treatments.

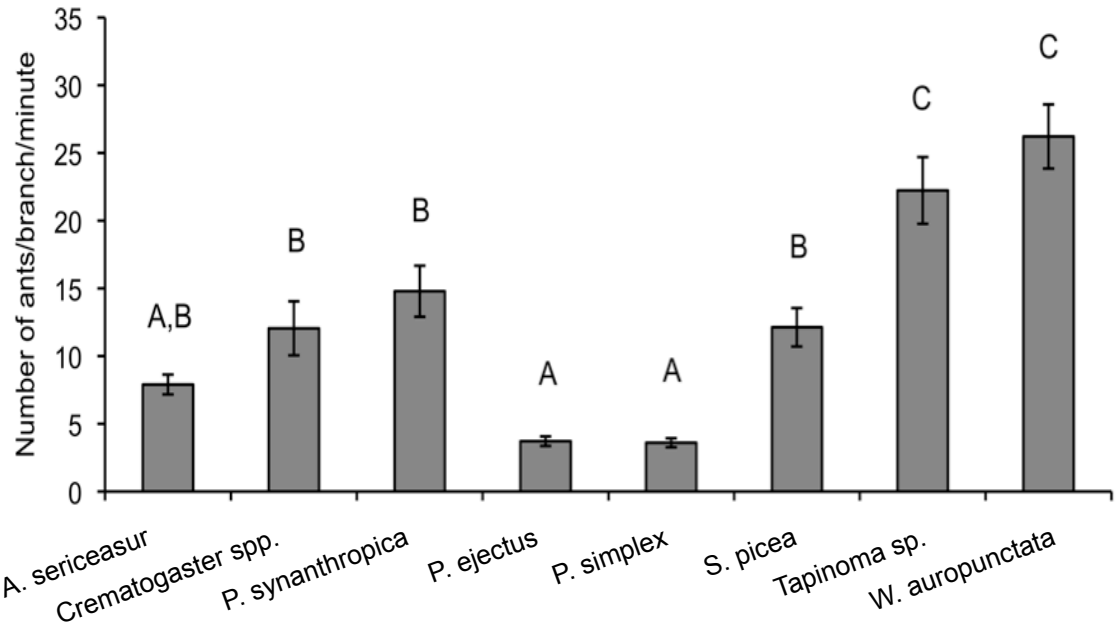

Figure 4.3. Means and standard errors of ant activity/branch/minute across the control branches of the ant species treatments. Common letters indicate means that are not significantly different from one another as determined by Tukey's HSD.

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Chapter 5
The pest suppression services of multiple ant-species are enhanced by herbivore diversity

Abstract

Increased prey diversity is hypothesized to limit the efficiency of single predator species relative to multiple predator species in suppressing prey communities. In most food webs, predators encounter diverse prey communities; however the majority of predator biodiversity experiments focus on the suppression of just a single prey species. Here, we compared the efficiency of multiple and single ant species at providing pest suppression of simple (single species) and diverse (three species) herbivore communities in coffee agroecosystems. Our results suggest that multiple ant species are more efficient at preventing diverse herbivore communities from damaging coffee compared to single ant species. However, single ant species were equally successful at suppressing single herbivore species relative to multiple ant species. These results support the hypothesis that greater prey diversity limits the success of single predators and that to suppress entire herbivore communities, greater diversity of predators may be needed. Further, these results have important implications for understanding the cascading effects of predator biodiversity on herbivores and plant communities that are especially relevant in agriculture where management for the control of entire pest communities is a primary goal.

Introduction

Effects of multiple predator species on prey abundance are extremely variable in comparison to other biodiversity-ecosystem function relationships. This variability is due to the potential for negative interactions among predators such as intraguild predation, interference competition, as well as niche partitioning and facilitation within the predator community (Sih et al. 1998, Letourneau et al. 2009, Griffin et al. 2013). A recent review of the experimental literature

suggests that although multiple predator species are more efficient than the average single predator species at suppressing prey, they are only as efficient as the most dominant single predator species (Cardinale et al. 2006, Griffin et al. 2013). This implies that conserving the most efficient predator should maintain ecosystem function, and is a result that has major implications for biological control of agricultural pests and invasive species.

However, most predator biodiversity experiments do not specifically test if multiple predators limit prey communities and do not look at the resulting cascading effects on lower trophic levels (i.e. primary producers). The majority of multiple predator experiments focus on just one prey species alone and just 23% (21/93) of experiments incorporate more than two prey species (Cardinale et al. 2006, Griffin et al. 2013). For agricultural ecosystems, these statistics are even narrower, where 64% (38/59) of studies focus on one prey (herbivore) species and just one experiment out of 59 included more than two prey species. Further, 93% of experiments focused on aphid species only, while the remaining four investigated leafhoppers or planthoppers (other families within the order Hemiptera), suggesting substantial taxonomic bias. These statistics represent a major shortcoming of the current literature as very few studies experimentally test the importance of predator biodiversity on entire prey communities. Even simple agroecosystems, such as corn or soy fields, may contain diverse herbivore communities of more than 20 species (O'Day and Steffey 1998, Herbert and Malone 2012). Thus, to understand the importance of predator biodiversity on ecosystem function, more community-wide experiments or at minimum, the inclusion of more prey species within experiments is needed.

There are several theoretical reasons why the effects of multiple predator species on single prey species may differ from their effects on the entire prey communities (Wilby and Thomas 2002, Briggs and Borer 2005, Tirok and Gaedke 2010, Tylianakis and Romo 2010). For instance, increasing prey species diversity should lead to a greater resource niche space for predator species, which may lead to greater niche partitioning. Single predator species may be dominant in capturing prey of one or several functional prey types, however, increasing prey diversity may introduce prey types that a given predator is ill-equipped to consume. Indeed, a review of the multiple predator experiments in marine and aquatic ecosystems suggests that in predator experiments, increased prey diversity on average limits the success of predators (Edwards et al.

2010). However, no study has demonstrated how multiple predator species are more efficient at suppressing diverse prey communities compared to single predator species. In this study, we use laboratory experiments to explore the importance of single- and three-species ant communities in suppressing damage to coffee (*Coffea arabicia* L.) by single and three-species herbivore communities. As we show next, single ant species were equally efficient at suppressing each single herbivore species relative to the multiple ant species. However, multiple ant species were more efficient at suppressing the multiple herbivore treatment relative to single ant species.

Methods

We conducted research on coffee plantations in the Soconusco region of Chiapas, Mexico in March-July of 2013. Neotropical coffee agroecosystems may host upwards of 200 species of arthropod herbivores of coffee (Le Pelley 1973). We focused our study on three herbivore species. The small (<1mm) coffee berry borer (*Hypothenemus hampei* Ferrari, 1867 [Coleoptera: Curculionidae]) is the most important arthropod pest because adults bore into coffee berries, lay eggs, and the larvae damage the harvestable seeds (Damon 2000). The adults of *Rhabdopterus jansoni* (Jacoby) (Coleoptera: Chrysomelidae) chew coffee leaves and the fleshy outer parchment of berries, but rarely cause economic damage (Barrera et al. 2008, Kuesel et al. 2014). *Macunolla ventralis* (Signoret 1854a: 21; [Hemiptera: Cicadellidae]) is a common polyphagous leafhopper that feeds from the xylem of coffee. Although leafhopper damage to coffee is minor, some Cicadellidae are vectors of important coffee pathogens (Redak et al. 2004). Coffee-ecosystems also host a diversity of predators (Perfecto et al. 2007). We focused on three ant species that suppress pests via consumptive and non-consumptive effects; ants actively remove herbivores, but do not always consume them. *Azteca sericeasur* Longino was previously referred to as *A. instabilis*, but has been re-identified as *A. sericeasur* due in part to the queens' smaller ocelli and distinct yellow and brown facial markings (J. Longino, *pers. comm.*). *Azteca sericeasur* is a dominant, arboreal species that dictates the structure of the food web on coffee (Vandermeer et al. 2010). *Pseudomyrmex ejectus* F. Smith 1858 and *Pseudomyrmex simplex* F. Smith 1877 are arboreal ants that nest in hollowed coffee branches. These three species suppress *H. hampei* damage (Gonthier et al. 2013), but their effects on other herbivores are unknown.

To determine if the effects of multiple ant species on herbivores are mediated by herbivore diversity, we conducted laboratory experiments at Finca Irlanda using insect arenas (60×60×60cm; Bug Dorm-2, Bug Dorm Store, Taichung, Taiwan) following methods modified from Philpott et al. (2012). We introduced coffee branches, herbivores (1 sp. or 3 spp.), and predators (0, 1 sp., or 3 spp.) into arenas for a 4×5 factorial experiment (Table 5.1). Two coffee branches, tied together at the stem, were introduced to each arena. To eliminate pre-existing damage of coffee, the first branch contained 4-8 young, undamaged leaves with no berries and the second branch contained 10 un-ripened, undamaged berries with no leaves. Herbivore treatments followed an additive design because of the size disparity between species. Single species treatments were as follows: (1) for *H. hampei,* we released 20 adult individuals, (2) for *M. ventralis* we released 3 individuals, and (3) for *R. jansoni* we released 3 individuals. We released the same numbers of each prey species in the three-herbivore species treatment. Ant species treatments compared no ants, 1 ant species (*A. sericeasur*, *P. ejectus,* or *P. simplex*), or 3-ant species treatments with a substitutive design. We held ant worker density at approximately 36 workers across the 1 species and 3 species (12 workers of each species) treatments, however we also incorporated *P. ejectus* and *P. simplex* brood and twig nest material to improve normal worker activity. We collected *M. ventralis* with sweep-nets and *H. hampei* were removed from dry, old infested coffee berries no later than 1h before the initiation of experiments because these species are sensitive in captivity. We starved *R. jansoni* for 24h to improve feeding activity. *Azteca sericeasur* workers were collected by hand from nearby nests and *P. ejectus* and *P. simplex* nests were collected from destructive sampling of their nests (dry, hollow coffee twigs) no longer than 24h before experiments.

To quantify herbivore damage after the 24h experimental duration, we counted the number of coffee berries infested by *H. hampei* and counted the cm^2 of leaf tissue damage by *R. jansoni.* However, because the stylet-feeding damage of *M. ventralis* is difficult to measure, we measured the presence of *M. ventralis* on coffee plant tissue as a proxy for damage. Laboratory observations suggested that 77% (20/26) of *M. ventralis* individuals placed on coffee leaves fed within 20 minutes. At time 0.5h, 6h, and 17h from the experimental initiation, we counted *M. ventralis* individuals on coffee and calculated the average number of *M. ventralis* per experimental replicate. For the 3-herbivore species treatment, we measured the three damage

types and created a 'damage index' following methods used in the multi-functionality literature (Zavaleta et al. 2010, Maestre et al. 2012). We standardized each damage type measurement by calculating the z-score ($x - \text{mean}_x/\text{standard deviation}_x$) and averaged the scores of each damage type as in Maestre et al. (2012). We favored z-score transformations over other methods (Zavaleta et al. 2010) because it followed a normal distribution and did not limit data variability to values between 0 and 1 (Maestre et al. 2012).

To evaluate the effect of ant biodiversity on suppression of herbivore communities with 1 or 3 species, we conducted two sets of analyses. First, to determine the 'average effect of ant diversity' (model 1), we compared the suppression of herbivore damage in the treatment with 3-ant species, the average of the treatments with 1 ant species, and the treatment with no ants with generalized linear models (GLM). To do so, we conducted a separate analysis for each herbivore species alone and the treatment with all 3-herbivores species alone. We assessed the distribution of each measurement of damage with Kolmogorov-Smirnoff tests and qq-plots. In all models a Gaussian error distribution was assumed, except for damage by *R. jansoni,* which fit a negative binomial distribution with a log-link function. In this first analysis (model 1), we incorporated the 'average effect of ant diversity' (no predator control, average 1 species treatment, and 3 species treatment) into models as a fixed effect. We calculated pair-wise comparisons among groups with estimated marginal means (t-tests) to distinguish levels within factors (Post hoc test). Our second analysis (model 2) was aimed at evaluate the effect of 'ant species identity' and if the treatment with 3-ant species suppressed pests better than the most efficient treatment with 1 ant species. The effect of species identity (model 2) was evaluated by comparing the pest suppression of the 4 herbivore treatments by the three ant species, *A. sericeasur, P. ejectus,* and *P. simplex*. As above, we ran GLMs assuming Gaussian error distribution, except for the *R. jansoni* model, which was fit with negative binomial distribution. We also calculated pair-wise comparison among groups with estimated marginal means (t-tests) to distinguish levels within the factor (Post hoc test).

Results

The analyses of the average effect of ant species diversity (model 1) revealed that treatments with 3-ant species had no difference in pest suppression relative to the average of the treatments with 1 ant species for each of the individual herbivore treatments; *M. ventralis, H. hampei,* and *R.*

jansoni (Table 5.2, Fig. 5.1A,B,C; see Fig. 5.1 for post hoc comparisons). In contrast, in the treatment with 3 herbivore species, the treatment with 3 ant species had a 40% lower herbivore damage index than the average of the treatments with 1 ant species (Fig. 5.1D). In each of the 1 herbivore species treatments, the average of the treatments with 1 ant species and the treatment with 3 ant species both had lower herbivore damage than the no ant treatment. In the treatments with 3 herbivore species, although the treatment with 3 ant species had greater pest suppression than did the treatment with no ant species, the average of the treatments with 1 ant species did not differ in pest suppression relative to the treatment with no ant species.

The species identity analyses (model 2) revealed that across each of the treatments with 1 herbivore species, the treatment with 3 ant species was only more efficient than *P. ejectus* at suppressing *M. ventralis* and *H. hampei* herbivores (Table 5.2, Fig. 5.1E,F). In the *R. jansoni* herbivore treatment, there was no difference between any of the treatments with 1 ant species and the treatment with 3 ant species (Table 5.2, Fig. 5.1G). In the treatment with 3 herbivore species, the treatment the 3 ant species had significantly lower damage scores than the treatments with *P. ejectus* (50% lower)*, P. simplex* (42% lower)*,* and *A. sericeasur* (26% lower) ant species (Fig. 5.1H). Of the treatments with 1 ant species, *A. sericeasur* typically provided the most efficient pest suppression and was even marginally better than the treatment with 3 ant species at suppressing *M. ventralis* and *H. hampei* alone. The treatment with *A. sericeasur* significantly suppressed herbivores relative to the no ant treatment in all four herbivore treatments. The treatment with *P. simplex* had lower damage by *H. hampei* compared to the no ant treatment. However, in all other herbivore treatments, treatments with *P. simplex* and *P. ejectus* ant were no different than the no ant treatments.

Discussion

This is the first study, to the knowledge of the authors, which supports the hypothesis that herbivore diversity enhances the efficiency of multiple predators relative to single predators. When only one herbivore species was considered, treatments with 1 ant species typically provided equal pest control relative to the treatment with 3 ant species. However the treatment with 3 ant species was better at suppressing the three herbivores simultaneously. The treatment with 3 ant species was also more efficient at suppressing the diverse herbivore community

relative to the most efficient ant species (*A. sericeasur,* Fig. 4.1D). While this might suggest multiple ant species exhibit "transgressive overyielding," this analysis is suggested to be a statistically biased metric and no longer the best method for interpreting biodiversity effect mechanisms (Schmid et al. 2008). Unfortunately, other methods, such as, additive partitioning models (Loreau et al. 2001, Saleem et al. 2012) are not feasible in predator-prey studies because the contribution of each predator species to prey suppression in polyculture is very difficult to measure. Thus, the relative importance of selection effects and complementarity are unknown for this multiple predator experiment.

To date, few studies have tested if greater prey diversity limits the efficacy of single predator species relative to multiple predators. Wilby and Orwin (2013) compared pest suppression efficiency of multiple and single predators in treatments of 1, 2, or 4 aphid species, but found that multiple-predator effects were weakened by increased herbivore species richness. Douglass et al. (2008) revealed grazer richness increased predator resistance in multiple predator species treatments, but did not affect single predator species effects in a marine benthic community. Saleem et al. (2012) found similar results, where protist predator diversity effects were weaker when bacteria prey diversity was higher. Snyder et al. (2008) showed multiple predators always outperformed single predators at controlling 1 or 2 aphid species. Thus, there has been no clear pattern across the experiments that simultaneously manipulated prey and predator diversity within study designs.

If each plant species represented the base of a food web, the typical terrestrial food web would contain a diverse herbivore community (Price 2002, Novotny and Basset 2005, Gilbert et al. 2012), yet the majority of multiple predator experiments focus on single prey species (Cardinale et al. 2006, Griffin et al. 2013). It could be argued that the literature has failed to capture the effect of predator diversity on the abundance and biomass of prey communities or the resulting trophic cascades on lower primary producer trophic levels. Specifically, these findings have major implications for agricultural ecosystems. While the focus of much natural and chemical pest control research centers on the control of single, important pest species, agroecosystems typically contain a great diversity of pest species and few studies have considered how diverse assemblages of predators are important to regulating the multiple pests simultaneously. Our

results support the notion that diverse pest communities require diverse predator communities to maintain pest communities at lower thresholds.

There is now a vast amount of literature that suggests incorporating other forms of food web complexity into predator biodiversity experiments reveals context-dependent effects of biodiversity on ecosystem function (Duffy et al. 2005, Duffy et al. 2007, Douglass et al. 2008, Srivastava and Bell 2009, Philpott et al. 2012, Wilby and Orwin 2013). However, increasing diversity at multiple trophic levels and modifying food-chain length has also revealed complicated and inconsistent impacts on the effects of biodiversity at different trophic levels. Nonetheless, these experiments highlight that the effects of biodiversity are not always describable by simple experimental designs, and therefore incorporating more complexity and realism in experiments should help reveal the true importance of biodiversity. This study supports the hypothesis that the suppression of herbivores by multiple predator species is enhanced relative to single predator species when the herbivore community is diverse, but not when the herbivore community is simple. Given that the majority of multiple predator experiments have focused on one prey species, our results suggest that considering more prey species within experiments may reveal more accurate effects of multiple predator species within ecosystems.

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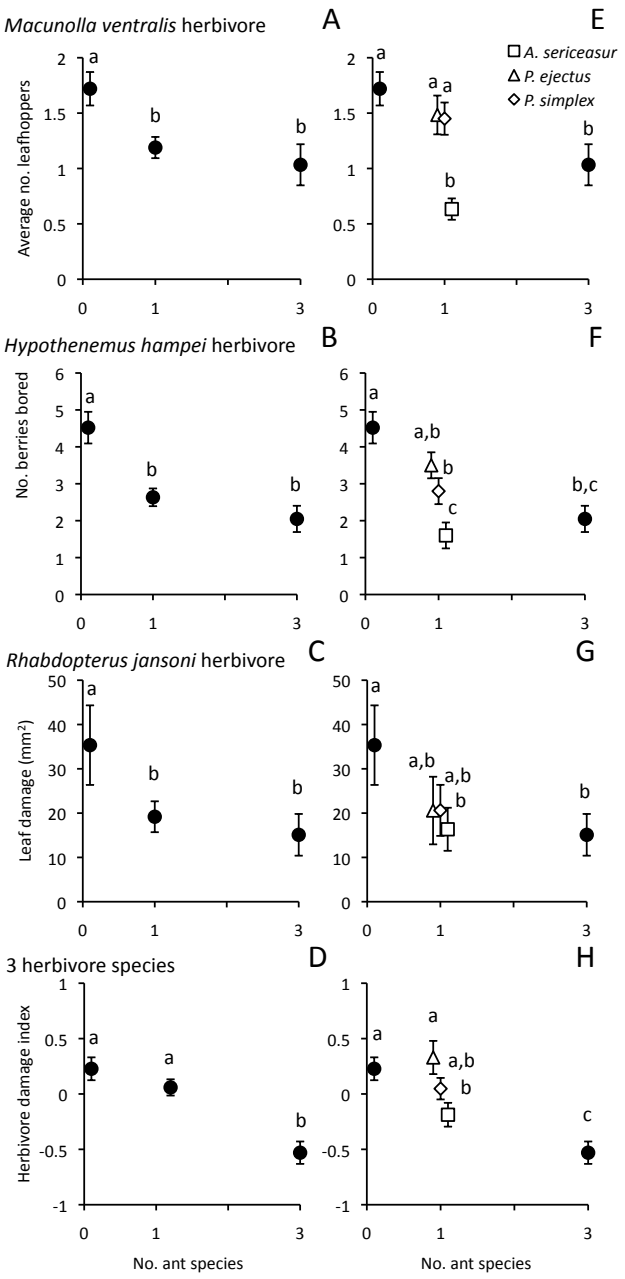

Figure 5.1. Mean (±SE) herbivore damage estimation across treatments with no ant species, 1 ant species, and 3 ant species. Average effects of 1 ant species are reported in panels A-D and the effects for each ant species are reported in panels E-H. Herbivore treatments included: *M. ventralis* leafhoppers on coffee branches (A, E), *H. hampei* infesting coffee berries (B, F), leaf damage (cm^2) from *R. jansoni* leaf beetles (C, G), and the herbivore damage index of treatment with all 3 herbivores (D, H). The herbivore damage index was defined by the average z-score transformed damage measurement for each herbivore species. Black symbols represent the means for treatments with 0, 1, or 3 ant species. Letters represent statistically ($p<0.05$) different levels between treatments, as determined by post hoc tests.

Table 5.1. Experimental treatments and sample size.

Predator	Herbivore	N
No predator	Total	97
	H. hampei	25
	M. ventralis	25
	R. jansoni	29
	3 herbivore spp.	18
Single predator	Total	240
A. sericeasur	*H. hampei*	20
	M. ventralis	20
	R. jansoni	20
	3 herbivore spp.	21
P. ejectus	*H. hampei*	20
	M. ventralis	20
	R. jansoni	19
	3 herbivore spp.	20
P. simplex	*H. hampei*	20
	M. ventralis	20
	R. jansoni	20
	3 herbivore spp.	20
Three predator	Total	81
	H. hampei	21
	M. ventralis	20
	R. jansoni	20
	3 herbivore spp.	20

Table 5.2: Statistical comparisons predator and herbivore diversity experiment.

	df	F	P
H. hampei			
M1: Ave. species effect	2,103	11.9	<0.001
M2: Identity effect	4,101	9.3	<0.001
M. ventralis			
M1: Ave. species effect	2,102	5.6	0.005
M2: Identity effect	4,100	7.9	<0.001
R. jansoni			
M1: Ave. species effect	2	10.1*	0.007
M2: Identity effect	4	10.8*	0.029
3-herbivore spp.			
M1: Ave. species effect	2,96	11.9	<0.001
M2: Identity effect	4,94	9.1	<0.001

*Wald χ^2 statistics; M1 = Model 1 average species effects; M2 = Model 2 species identity effects

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Chapter 6
Context-dependent interaction cascades in Mexican coffee

Abstract

Complex ecological communities are composed of long chains of directly and indirectly interacting species that maintain ecosystem properties. The strength, sign, and length of these links between species also depend on the biotic and abiotic environmental context surrounding the community. In agricultural ecosystems the management for one beneficial species interaction may therefore have profound effects on the interactions among other species and other ecosystem services. Here, using observational and experimental studies from Mexican coffee plantations, we evaluate the ecosystem services provided by shade trees and the arboreal ant *Azteca sericeasur* that nests in shade trees. We show that coffee bushes around shade trees in the genus *Inga* (containing nitrogen fixing bacterial associations) had greater yield than coffee bushes around non-*Inga* species trees. However, coffee bushes around non-*Inga* trees with *A. sericeasur* ants had 54% fewer coffee berries with berry borer pest damage and 28% lower proportion of leaves with leaf rust lesions relative to coffee bushes around *Inga* trees with ants. Lower proportions of leaves with leaf rust lesions negatively correlated with the abundance of green scales infected by the white halo fungus (a hyperparasite of the leaf rust), and green scales infected by the white halo fungus were also 29 times more abundant on coffee bushes around non-*Inga* trees relative to coffee around *Inga* trees with *A. sericeasur* ants. These results suggest that shade tree species (*Inga* and non-*Inga* species) modifies the pest and disease suppression services provided by *A. sericeasur* ants. Further, the greater pest and disease suppression services provided by *A. sericeasur* ants on non-*Inga* trees trades off with the soil enrichment services provided by *Inga* tree species.

Introduction

Complex ecological networks may sometimes make it difficult to understand the effect of one species on another species in predator-prey interactions, mutualistic interactions, competitive interactions, and other one-on-one interactions (Hairston et al. 1960, Paine 1966, Bronstein 1994, Yodzis 2000, Duffy et al. 2007, Pringle et al. 2007, Tylianakis and Romo 2010, McCauley et al. 2012). However, in many ecosystems, species interact with multiple species in many different ways. These interactions form large networks that indirectly link species together via long chains of direct interactions (indirect interactions). While some have assumed that distant indirect interactions between species are probably weak and therefore unimportant (Berlow et al. 2009), much empirical work has described the importance of many indirect effects (Paine 1966, Schmitz et al. 2000, Letourneau et al. 2004, Pringle et al. 2007, McCauley et al. 2012). Indeed, advances in network analysis suggest that considering additional existing trophic links within networks increases the predictability of one-on-one species effects (Berlow et al. 2009). Further the strength and sign of interactions between species are also context dependent on the biotic and abiotic environmental surrounding the community (Bronstein 1994, Duffy et al. 2007, Tylianakis and Romo 2010, Pringle et al. 2013).

In agricultural ecosystems, the set of species interactions within ecological networks are sometimes valuable (e.g., ecosystem services) (Daily 1997). Like other fields (e.g. medicine, economics, social science), the importance of complexity in agricultural science was acknowledged long ago, but technological advances and mechanization of production has often put agricultural complexity at odds with conventional agricultural practices (Howard 1940, Altieri 1987, Lewis et al. 1997, Tscharntke et al. 2005, Gliessman 2007, Power 2010, Vandermeer et al. 2010, Kremen et al. 2012). More recently many agricultural researchers have acknowledged that the production of crops requires favorable conditions with water, nutrients, and light and requires protection against competitors, pests, and pathogens (Foley et al. 2005, Power 2010, Snapp et al. 2010, Kremen et al. 2012, Iverson et al., in prep.). Humans may create these conditions synthetically. However, ecological communities may be responsible for some or all of the soil enrichment, weed control, pest control, and disease control services in some agricultural systems. Understanding how to manage agricultural ecosystems to maximize all services is thus an important goal, but trade-offs between services may exist (Foley et al. 2005, Power 2010, Kremen et al. 2012, Iverson et al., in prep.). Still very few studies have examined

how multiple services are provided by species simultanuously. Here we study how changes in non-crop species identity in coffee plantations modify how multiple ecosystem services are provided to coffee production.

Coffee is one of the most important tropical crops because it provides income for the livelihoods of millions of farmers and farm workers and has a high retail value (Pendergast 1999). In much of Latin America, coffee management varies greatly and traditionally it was grown in the understory of native diverse shade trees that provided a forest-like habitat (Perfecto et al. 1996). However, recently the region has transitioned toward simplification of the shade tree communities by heavily pruning trees and planting fewer species mostly in the genus *Inga* (Fabaceae) or removing trees altogether (Moguel and Toledo 1999, Jha et al. 2014). In addition to changing management, coffee has experienced a variety of other stressors like pests and disease. A number of economically important pests attack coffee, including the coffee berry borer (*Hypothenemus hampei*), which infests on average 6.6% of harvestable coffee fruits and in patches can reach up to 51% (data presented below). Even more recently, in late 2012 and early 2013, an epidemic of the coffee leaf rust (*Hemileia vastatrix*) resulted in major defoliation of coffee (60% of plants had 80% leaf loss in the study site) and it is estimated that up to 40-50% of yield was lost to coffee production throughout Latin America (Cressey 2013, Vandermeer et al. 2014).

In the Soconusco Region of Chiapas, Mexico, 14 years of investigations describe an interaction web that provides suppression of several pests (Vandermeer et al. 2010). The dominant, arboreal ant, *Azteca sericeasur* (previously referred to as *A. instabilis*), forms carton nests in shade trees where it has consumptive and/or non-consumptive effects on the berry borer, Lepidoptera larvae, leafhoppers, and a leaf-chewing beetle (*Rhabdopterus jansoni*) (Vandermeer et al. 2002, Perfecto and Vandermeer 2006a, Philpott et al. 2012, Gonthier et al., in prep.). Yet, *A. sericeasur* also forms a mutualism with the honeydew-producing hemipteran, *Coccus viridis* (coffee green scale), causing elevated populations of scales on coffee bushes near shade trees with ant colonies (Vandermeer and Perfecto 2006). The white halo fungus, *Lecanicillium lecanii,* exhibits epizootic outbreaks in large green scale patches limiting scale population size. Additionally, the white halo fungus is a hyper-parasite of the coffee leaf rust (*Hemileia vastatrix*) and outbreaks in green scale patches are correlated with the suppression of the leaf rust in the following seasons (Jackson et al. 2012). The *Azteca*-interaction web with coffee, *A. sericeasur*

ants, the coffee green scales, the berry borer, the white halo fungus, and the coffee leaf rust has been previously described in great detail in Vandermeer et al. (2010; Fig. 6.1).

The planting of mostly *Inga* spp. shade trees is very common within plantations in Mexico, Colombia and elsewhere because many *Inga* spp. host nitrogen-fixing bacteria and may improve soil quality (Dommergues 1987). However, this promotion of *Inga* trees may have unforeseen consequences for the efficiency of the *Azteca*-interaction web because *Inga* species' tree canopies harbor two significant resources for ants: large extrafloral nectaries on their leaves and high densities of honeydew-producing hemipterans in their canopies (Livingston et al. 2008). These canopy resources drive *A. sericeasur* foraging into tree canopies, and subsequently reduce ant foraging and the size of green scale populations on coffee bushes around shade trees (Livingston et al. 2008). Regardless, it is unknown how shade tree species effects on ant foraging modifies the suppression of the berry borer and the leaf rust. In this study, we used observational and experimental studies from a series of sites in coffee agroecosystems to determine the impacts of *Inga* and non-*Inga* trees on pest and disease suppression by the *Azteca*-interaction web. We also determined if soil enrichment services provided by *Inga* trees traded off with better pest and disease suppression by the *Azteca*-interaction web in coffee near non-*Inga* trees with *A. sericeasur* colonies.

Methods

1. Survey of Azteca sericeasur colonies in Inga and non-Inga trees

This study was conducted in four coffee production systems, Finca Irlanda production, Finca Irlanda restoration, Finca Hamburgo, and Finca Santa Anita, in the Soconusco region of Chiapas, Mexico. In May of 2011 (rainy season), we surveyed each production system and established four site locations within each plantation, except Finca Irlanda restoration, where we were limited to three sites because of the small size of the plantation. Within each site, we surveyed all trees for *A. sericeasur* colonies within a ~1-ha area and selected two occupied trees (one-*Inga* and one-non-*Inga*) separated by >20m. We chose only medium to large colonies and assumed each occupied tree represented a different ant colony. If similar sized colonizes were unavailable within a site, we searched nearby areas until we obtained two comparable colonies. In February (dry season) and May (rainy season) of 2012, we re-surveyed sites and searched for additional colonies if the trees hosting ant colonies had been heavily managed or removed, if ant colonies

had moved, or local management conditions had drastically changed (i.e., removal or replanting of coffee plants). The *Inga* tree species included within the study were, *I. micheliana*, *I. rodrigueziana,* and *I. vera*. The non-*Inga* tree species included in the study were, *Miconia affinis* (Melastomataceae), *Alchornea latifolia* (Euphorbiaceae), *Trema micrantha* (Cannabaceae), *Yucca elephantipes* (Asparagaceae), *Syzygium jambos* (Myrtaceae)*, Cordia stellifera* (Boraginaceae)*, Ficus* sp. (Moraceae), and *Ocotea* sp. (Lauraceae).

To determine how pest and disease suppression differed on coffee around *Inga* and non-*Inga* trees with and without *A. sericeasur* colonies, we opted for an observational survey instead of an exclosure or ant-removal experiment for several reasons. First, *A. sericeasur* has a patchy distribution with clear presence and absence in shade trees and nearby coffee making absence equivalent to control. Second, exclosure experiments with *A. sericeasur* at the study site suggest that the sticky residues (tanglefoot) used to exclude ants also exclude other crawling non-ant predators. For instance, sticky residues exclude coccinellid larvae that are important predators of the coffee green scale (Gonthier et al. 2013b). Previous research suggests exclusions can result in un-realistic patterns of high densities of green scales on ant-excluded coffee bushes (Philpott et al. 2008, Gonthier et al. 2013b). Finally, *A. sericeasur* removal was attempted via poisoning and carton-nest destruction over the course of a three-month period, however it was largely unsuccessful. Nests of *A. sericeasur* often penetrate deep into tree cavities and are not easily removed or destroyed (Personal observations K. Ennis). Even if *A. sericeasur* removal were successful, the great effort needed to do so would substantially limit overall sample size. Thus, we opted to compare the presence and absence of *A. sericeasur* in *Inga* and non-*Inga* species trees.

1.i. Colony and site description

At each shade tree with an *A. sericeasur* colony, we measured colony and site level factors. As a proxy for colony size, we measured a 'colony activity index,' by beating the trunk of the tree with wood poles and videotaping ant activity on three index cards (7.6×12.7cm) pinned to areas with high ant activity (Table 6.1). We counted the number of ants per card at 10s and 20s of footage and averaged across all cards per colony. We also measured a number of site level variables, including coffee density near each tree with *A. sericeasur* (within a 10×10m plot), the

number of trees in physical contact with the tree with *A. sericeasur* (hereafter nest-tree connections), and height of the tree with *A. sericeasur* (Table 6.1).

1.ii. Shade tree ant foraging and honeydew producers

For each tree with *A. sericeasur,* we selected seven visible and haphazardly chosen canopy branches, one from each of the four cardinal directions and at least one from the top of the canopy crown. For each branch, we used binocular or visual surveys to observe the number of ants crossing a point in one-minute (canopy ant activity; Table 6.1). To measure canopy honeydew producers, we cut down at least five haphazardly chosen branches, one from each of the cardinal directions and one from the top of the canopy crown. In all trees, sessile scale insects (Hemiptera: Coccidae), mealybugs (Pseudococcidae), and whiteflies (Aleyrodidae) were the most abundant honeydew producers. We standardized the number of honeydew producers per branch by the estimated total leaf area sampled. Study trees without *A. sericeasur* were not measured due to limitations of time.

1.iii. Coffee ant foraging

We recorded two measures of ant foraging on coffee bushes (Table 6.1). We observed the number of ants crossing a point on the main coffee trunk for one minute on three coffee bushes nearest to the base of the tree with *A. sericeasur* foraging (coffee ant activity). We also counted the number of coffee bushes (within 10x10 m plots surrounding the tree with *A. sericeasur*) with noticeable ant activity by shaking and beating the coffee trunks and observing ant movement (number of coffee bushes foraged).

1.iv. Coffee yield

To quantify yield, we counted the total number of coffee berries per bush on three coffee bushes near to shade trees with and without *A. sericeasur* (control trees). *Azteca sericeasur* foraged on between one and 25 bushes per colony, therefore we measured more bushes for those colonies that foraged on a greater number of bushes. In 2012, we also re-sampled sites counting the number of berries per bush for one bush at 1m, 4m, and 7m from the shade tree base. This extra sampling was aimed at gaining a better understanding the relationship between distance from shade trees and yield.

1.v. Coffee berry borer

1.v.a. Borer addition experiment

At each *Inga* and non-*Inga* tree with and with *A. sericeasur*, we located one coffee bush near to the base of the tree. We observed the coffee bushes without *A. sericeasur* for one minute to confirm that no other ant species was present. Then on one branch per coffee bush, we removed berries until each branch had exactly 15 berries all lacking borer entrance holes. We then added 20 berry borers to a leaf near to the berries on the branch. This enabled berry borers to get upright and locate the berries. After 24 hours, we counted the number of berries with borers. We also confirmed that coffee bushes around trees without *A. sericeasur* were devoid of ants by comparing berry colonization by the berry borer with sticky-residue excluded controls on the same plant. Coffee bushes around trees without *A. sericeasur* (mean±se; 5.0±1.2) did not differ from sticky-residue excluded controls (4.2±0.8), confirming that there was negligible ant activity on the coffee bushes selected around trees without *A. sericeasur* (paired t-test, t= -1.8, df=5, P=0.141).

1.v.b. Berry borer survey

In the rainy season of 2011-12, we measured the number of berries with borers on 3 bushes near to each focal tree across tree type and ant presence/absence. Ant colonies foraged on between one and 25 bushes, therefore we measured a higher number of bushes for colonies that foraged on a high number of bushes.

1.v.c. Ant colony-level effects on berry borer

To estimate the colony-level impact on the berry borer, we estimated the total number of berries saved from the borer per ant-colony. Because we found no relationship between the effect size of ants on the borer and distance from the nest, we assumed all bushes were protected equally. We calculated berries saved per colony (β) in a similar fashion to Karp et al. (2013) and Kellermann et al. (2008) as:

$\beta = \Delta$ borer*yield*bushes foraged

Where 'Δ borer' is the proportion of berries with borers in control (no *A. sericeasur*) bushes subtracted from the proportion of berries with borers in ant bushes, 'yield' is the average yield

per bush at the location, and 'bushes foraged' is the number of bushes on which ants were found foraging. These values were generated from means across the two years of the borer surveys (section 1.v.b.).

1.v.d. Valuation ant pest control of borer

Using the number of medium and large *A. sericeasur* colonies (α) in *Inga* and non-*Inga* trees per ha across 44 ha surveyed in nine coffee plantations in 2011-12, we estimated the value of *A. sericeasur* per ha as:

Value (US$ per ha) = $(\alpha_{Inga}*\beta_{Inga})+(\alpha_{non\text{-}Inga}*\beta_{non\text{-}Inga})*\phi*\omega$

Where β is the estimated berries saved per colony and α is the number of medium and large *A. sericeasur* colonies found in *Inga* or non-*Inga* trees, ϕ converts the number of berries to pounds (lb) of export quality un-roasted coffee or roasted coffee, and ω is the value (US$/lb) of export quality (International Coffee Association in 2011-12) or roasted coffee. The nine plantations surveyed each have their own processing plant and many sell export quality coffee and roasted coffee. Given that each plantation sells a range of coffee qualities that vary in price (ω; US$/lb), we provide a range of potential values of roasted coffee and export quality coffee that ranged from 1 to 10 US$ per lb.

1.vi. Coffee green scales & white halo fungus

In each season, we estimated the number of green scales on three coffee bushes at each study tree with *A. sericeasur* following methods from Vandermeer and Perfecto (2006). Study trees without *A. sericeasur* were not included due to limitations of time. First, we briefly counted the number of green scales per bush. If there were more than 6 scales per bush we used a four-class category estimation for each branch: 0-6 = low; 7-30 = medium; 30-70 = high; >70 = super high. During the scale counts, we also counted the number of green scales infected with the white halo fungus. If the number was greater than 6, we estimated the number of infected scales with a four-class category per branch: 0-5%; 5-15%; 16-35%; 35-75%; or >75% infected.

1.vii. Coffee leaf rust

In the dry season of 2013, we used the regional outbreak of the leaf rust to determine if there was a difference between the outbreak of leaf rust on coffee around *Inga* and non-*Inga* shade trees

with and without *A. sericeasur*. To calculate rust infection rate, we counted the total number of leaves and the number of leaves with rust lesions on 3 bushes per study tree, as in Avelino et al. (2012). On those same 3 coffee bushes, we also calculated the defoliation rate by counting the number of branches and the number of defoliated branches (lacking any leaves) per coffee bush.

2. Long-term data for green scales and white halo fungus

A 45-hectare plot within Finca Irlanda was established in 2004. At yearly or half-yearly intervals, each shade tree within the plot was surveyed for *A. sericeasur* ant colonies (Vandermeer et al. 2008). Starting in 2006, at each tree with *A. sericeasur*, the presence or absence of green scale populations and white halo fungus outbreaks in the surrounding coffee bushes were noted.

3.Analysis

For all analyses, we chose a repeated-measures Generalized Linear Mixed Model (GLMM) framework to determine how different variables responded to tree type (*Inga* and non-*Inga*) and season. We accounted for local-level differences by incorporating a site-level random effect. In each model, we included fixed effects of tree type, season, and their interaction. For dependent variables (yield, berry borer measurements, leaf rust measurements) that were measured on coffee around shade trees with and without *A. sericeasur* colonies, we also included the presence or absences of *A. sericeasur* in the model and all two- and three-way interactions. All analyses were conducted in SPSS (21.0); except for analysis of proportion data, which was analyzed using the 'lme4' package in the Program R (3.0.1).

3.i. Colony and nest-tree characteristics

We created GLMMs for the dependent variables nest-tree connections (Poisson), height (Gaussian), colony activity index (Gaussian), and coffee density (Gaussian). In each model, season, tree type, and the interaction between type and season were incorporated as fixed effects.

3.ii. Ant foraging and honeydew producers in nest-trees and coffee bushes

We compared the log of ant canopy activity (Gaussian), the number of honeydew producers per leaf area in trees with *A. sericeasur* colonies (Gaussian), the ant activity per coffee bush (Gaussian), and the number of coffee bushes foraged per colony (Gaussian), using GLMM with

the factorial comparison of season and tree type as fixed effects, colony activity index as a covariate, and the site as a random effect.

3.iii. Coffee yield

We compared the yield per bush (Gaussian) using GLMM with tree type, season, presence/absence of *A. sericeasur,* and all two- and three-way interactions included in the model. We also ran an additional GLMM (Gaussian) on data collected in 2012 that included distance from the shade tree (1, 4, 7m), presence/absence of *A. sericeasur,* and tree type as main effects and all two- and three-way interactions.

3.iv. Berry borer

For the borer addition experiment, we compared the number of berries with borers (after 24h) using GLMM (Poisson) with tree type and *A. sericeasur* presence/absence as fixed effects in the model. For the two year survey data, we compared the proportion of berries with borers using GLMM (binomial for proportion data), with season, tree type, *A. sericeasur* presence/absence, and all two-way and three-way interactions in the model. To determine colony-level effects of *A. sericeasur* on the berry borer, we compared the estimated number of berries saved with GLMM (Gaussian) with season, tree type, and their interaction as fixed effects with coffee bush density and colony activity index as covariates.

3.v. Green scales, white halo fungus, and leaf rust

Green scale abundance was compared using GLMM (negative binomial) with tree type and season and their interaction as fixed effects, colony activity index as a covariate, and site as a random effect. The number of green scales infected with the white halo fungus was compared using GLMM (negative binomial) with tree type and season and their interaction as fixed effects, colony activity index as a covariate, and site as a random effect. We chose to use the total number of white halo fungus infected scales rather than the proportion infected scales because the total number of infected scales is a measure that would be more relevant to the suppression of the leaf rust. We compared the proportion of defoliated branches and the proportion of leaves with leaf rust lesions using GLMM (binomial for proportion data) with tree type and ant presence/absence and their interaction as fixed effects (only dry season 2013).

3.vi. Long-term data for green scale and white halo fungus

We used a repeated-measures generalized linear models (GLM) with a binomial distribution to compare the probability of the presence of green scale populations and white halo fungus outbreaks in coffee around *A. sericeasur* colonies in *Inga* and non-*Inga* trees (fixed effect in model). Year was not included as a fixed effect in the statistical model due to differences in sampling effort between years.

Results

1. Two-year survey of A. sericeasur colonies

Azteca sericeasur was twice as active and tended 5.69 times more honeydew producers (per leaf area) in *Inga* relative to non-*Inga* tree canopies (Table 6.2; Table 6.3). In the 2012 rainy season, *A. sericeasur* activity was 2.4 times higher on coffee bushes around non-*Inga* trees relative to around *Inga* trees. However, in the 2011 rainy season, there was no difference in ant activity on coffee bushes (Table 6.2; Table 6.3). *Azteca sericeasur* ants from non-*Inga* colonies foraged on twice as many coffee bushes. The colony activity index did not significantly vary between colonies in *Inga* and non-*Inga* trees. *Inga* and non-*Inga* trees did not differ in the number of canopy connections, tree height, or coffee density (Table 6.2, Table 6.3).

2. Coffee yield

The presence of *A. sericeasur* did not impact the number of berries per coffee bush, however the number of berries in coffee bushes around *Inga* trees was between 1.7-1.8 times higher than coffee bushes around non-*Inga* trees (Table 6.4, Fig. 6.2A). Differences in the number of berries on coffee bushes around *Inga* and non-*Inga* trees were significant within 1m from trees (*post hoc* t-test; mean difference ± SE, 368±141, P=0.011), but not at 4m (117±141, P=0.409) or 7m (159±141, P=0.261) (Table 6.4, Fig. 6.2B).

3. Berry borer damage

The berry borer addition experiment revealed that coffee bushes around non-*Inga* trees with *A. sericeasur* had 69% fewer berries with borers relative to coffee around non-*Inga* trees without *A.*

sericeasur and 62% fewer berries with borers than did coffee around *Inga* trees with *A. sericeasur* (Table 6.5, Fig. 6.3A). However, the number of berries with borers did not differ between coffee bushes with and without *A. sericeasur* around *Inga* trees.

The survey results revealed an interactive effect of ant-presence and tree type on the percent of berries with borers. The presence of *A. sericeasur* reduced the percent of coffee berries with borers per by 67% around *Inga* trees and by 87% around non-*Inga* trees, compared to bushes without ants in each tree type (Table 6.5, Fig. 6.3B). Further, coffee around non-*Inga* trees with *A. sericeasur* had 54% fewer berries with borers than did coffee around *Inga* trees with *A. sericeasur*. On ant-free coffee bushes, non-*Inga* trees had 16% more berries with borers relative to *Inga* trees.

Azteca sericeasur in non-*Inga* trees saved an estimated 4.6 times more berries from borers relative to ants in *Inga* trees (Table 6.5, Fig. 6.3C). Although ant colony activity did not correlate with the number of berries saved, the coffee density positively correlated with the number of berries saved. *Azteca sericeasur* provided an estimated $0 and $55 (per ha) in pest suppression services. The mean value (per ha) of *A. sericeasur* pest suppression ranged from US$ 0.99 (per ha) to US$ 9.94 (per ha) depending on the price (per pound) of roasted or exported quality coffee (Fig. 6.3D).

4. Green scales, white halo fungus, leaf rust

Coffee bushes around non-*Inga* trees with *A. sericeasur* had 30.9 times more scales than coffee bushes around *Inga* trees with *A. sericeasur* (Table 6.6, Fig. 6.4A). Colony activity was positively, although only marginally, correlated with green scale density. There was no relationship between the number of green scales per bush and the yield per bush (parameter = 0.066±0.07, Wald's $\chi^2 = 1.1$, P=0.298), after distance to the nearest shade-tree and tree-type were taken into account in the model. Coffee bushes around non-*Inga* trees with *A. sericeasur* had 29 times greater densities of green scales infected with the white halo fungus relative to coffee bushes around *Inga* trees with *A. sericeasur* (Table 6.6, Fig. 6.4B).

In 2013, coffee bushes around non-*Inga* trees with *A. sericeasur* had 46% lower proportions of leaves with leaf rust compared to non-*Inga* trees without ants and had a 28.5% lower proportions than did coffee around *Inga* trees with *A. sericeasur* (Table 6.6, Fig. 6.4C). In contrast, coffee bushes around *Inga* trees with *A. sericeasur* had 20% higher proportions of leaves with leaf rust than bushes around *Inga* trees without *A. sericeasur.* Additional analysis revealed that there was a negative correlation between the number of scales infected with the white halo fungus in 2012 and the proportion of leaves with leaf rust lesions in 2013 ($Z=-11.5$, $p<0.001$).

The proportion of defoliated branches was 12% lower on coffee bushes around trees with *A. sericeasur* relative to bushes around trees without *A. sericeasur* (pooling both *Inga* and non-*Inga* trees) (Table 6.6, Fig. 6.4D). Further, coffee around non-*Inga* trees had 5.8% greater proportion of defoliated branches relative to coffee around *Inga* trees (pooling both trees with and without *A. sericeasur*) (Table 6.6, Fig. 6.4D).

5. Long-term data on the green scales and white halo fungus

The eight-year survey of the 45-ha plot in Finca Irlanda supported patterns found above for the green scale and white halo fungus. The frequency of encountering green scales was higher on coffee around non-*Inga* trees with *A. sericeasur* relative to on coffee around *Inga* trees with *A. sericeasur* (Fig. 6.5A, $F=13.3$, $df=1,2059$, $P<0.001$). Similarly, the frequency of encountering white halo fungus outbreaks was higher on coffee around non-*Inga* trees with *A. sericeasur* relative to coffee around *Inga* trees with *A. sericeasur* (Fig. 6.5B, $F=9.0$, $df=1,2059$, $P=0.003$).

Discussion

Our results suggest that different shade tree species that house *A. sericeasur* ant colonies may modify the strength of pest and pathogen suppression services provided by *A. sericeasur.* Further, the greater pest and pathogen suppression provided by ants nesting in non-*Inga* trees trades off against the greater soil enrichment services provide by *Inga* trees (Fig. 6.1). When nesting on non-*Inga* trees, *A. sericeasur* forage more on coffee bushes than when they nest on *Inga* trees. This is because *Inga* trees host high densities of honeydew producers and have extrafloral nectaries that provide sugar resources for ants and draw them into the canopy (Livingston et al.

2008). On non-*Inga* trees, in the absence of these canopy resources, *A. sericeasur* forages more on coffee. When on coffee, the ant reduces the berry borer, but increases green scale densities. Increased densities of green scales result in larger outbreaks of the white halo fungus. Because the white halo fungus also attacks the leaf rust, high densities of green scales infected with the white halo fungus may reduce the prevalence of the leaf rust (Jackson et al. 2012). Indeed, in the leaf rust outbreak of 2013, coffee around non-*Inga* trees with *A. sericeasur* had less leaf rust than did the coffee around *Inga* trees with *A. sericeasur.* However, the yield of coffee bushes was significantly lower near to non-*Inga* relative to *Inga* trees (in 2011 and 2012), regardless of the presence or absence of *A. sericeasur*. Given this trade-off in services, it is difficult to predict which tree type may be most beneficial for sustainable coffee production.

The number of berries per bush, as a measure of yield, was influence by the tree-type (*Inga* or non-*Inga*), but only at close distances to trees (Fig. 6.1B). There are several reasons that non-*Inga* trees might have lowered yields compared to *Inga* trees. First, non-*Inga* trees potentially have greater shading or crowding effects on bushes that are close by the base of the shade tree. Second, *Inga* trees are associated with nitrogen-fixing bacteria in root nodules and may have enriched and concentrated soil nitrogen around *Inga* crowns in close proximity to the shade tree trunk, countering any negative effects of *Inga* competition with coffee (Dommergues 1987). Both of these factors may have simultaneously contributed to differences between coffee yields on bushes near to *Inga* and non-*Inga* trees.

As speculated by (Livingston et al. 2008), activity of *A. sericeasur* was higher in the canopy of *Inga* trees compared to non-*Inga* trees resulting in lower activity of ants on coffee around *Inga* trees compared to non-*Inga* trees (Table 6.2). This pattern is likely driven by greater hemipteran and nectar resources in the canopies of *Inga* trees compared to non-*Inga* trees. In other systems, these effects are described as competition between honeydew-producing hemipteran species for ant attendance. Experimentally increasing the density of honeydew-producers on neighbor plants decreases ant abundance, membracid abundance, and increases spider predator abundance on focal plants (Cushman and Whitham 1991). Hence, the benefits of ants to hemipterans are lost in the presence of other resources. In our study, we show that the density of sugar resources (hemipterans and extrafloral nectaries) on plant neighbors correlates with reductions of (1)

honeydew-producers on the focal crop (coffee), (2) borer pest abundance, and (3) crop pathogen prevalence.

Greater ant foraging led to greater densities of green scales, however there was no negative correlation of the density of green scale on yield per bush, suggesting that green scales minimally impact coffee yield. These findings are somewhat surprising considering other studies have shown that in laboratory conditions, coffee seedlings may decline in growth rate with increasing infestation of green scales (Lemes Fernandes 2007). Field conditions vary from bush to bush and this variation may have many potential differences that mask negative effects of scales on coffee yield. For one, rapid population growth of green scales is often quickly countered by large outbreaks of the white halo fungus in the study region. If green scale densities only reach damaging levels for a short period than coffee may not experience significant yield losses.

The outbreaks of the white halo fungus in sites with high densities of green scales in 2012 were correlated with lower leaf rust prevalence in the dry season of 2013. Coffee around non-*Inga* trees with *A. sericeasur* had more green scales infected with the white halo fungus and a lower proportion of leaves with rust lesions than did coffee around *Inga* trees with *A. sericeasur.* These findings are similar to those in Jackson et al. (2012) where the density of the white halo fungus in one season negatively correlated with the lower densities of leaf rust in a second season.

Theoretical descriptions of the green scale, white halo fungus, and coffee leaf rust interactions suggest that coffee systems may exist in an alternative regime scenario whereby a high carrying capacity of the white halo fungus will lead to stable equilibrium of the leaf rust disease at low densities (Vandermeer et al. 2014). On the other hand, changes in management that lower the carrying capacity of the white halo fungus will lead to more complex dynamics and multiple equilibrium points some at high densities of the leaf rust (Vandermeer et al. 2014). The take-way message for managers is to consider managements that raise the carrying capacity of the white halo fungus. Vandermeer et al. (2014) describe how the density of *A. sericeasur* colonies is negatively correlated with the percentage of leaf rust damage on coffee. Our results add to this discussion by encouraging the planting of shade trees other than *Inga* species because when *A.*

sericeasur nests in non-*Inga* species it increases the size of epizootic outbreaks of the white halo fungus.

The increased ant foraging on coffee around non-*Inga* trees resulted in greater pest control of the berry borer. These results corroborate other studies on coffee that suggest *A. sericeasur* impacts borer colonization of berries (Perfecto and Vandermeer 2006b, Philpott et al. 2012, Gonthier et al. 2013a). Further, they suggest that the indirect effects of shade trees on ant foraging indirectly benefits coffee. It should be noted that for trees without *A. sericeasur,* it appears that coffee around *Inga* trees had fewer berries bored than did coffee around non-*Inga* trees. It could be that non-*Inga* trees happened to be in locations with greater densities of the berry borer. Another explanation might be provided by a recent study that suggests that *Inga* tree extrafloral nectaries promote natural enemies of coffee pests resulting in increased pest control near to *Inga* tree bases (Rezende et al. 2014). Indeed, Rezende et al. (2014) show that the number of ant and parasitoid visitors to extrafloral nectaries on *Inga* trees correlated positively with the number of parasitized leaf miners. In our study system, *Inga* trees might facilitate other natural enemies of the berry borer, parasitoids and other ant species (Gonthier et al. 2013a), and provide coffee near to *Inga* trees (without *A. sericeasur*) with facilitated pest suppression.

It is important to provide farmers with a translation of ecological analysis into economic terms (Daily 1997). In our study, we provide an analysis of the value of *A. sericeasur* suppression of the berry borer. However, we show that *A. sericeasur* also impacts the leaf rust defoliation indirectly in years of rust outbreak (Fig. 4CD) and we know *A. sericeasur* ants suppress other pest species in the system (Vandermeer et al. 2002, Gonthier et al., in prep.). Therefore our estimate of *A. sericeasur*'s value to coffee production could be considered conservative. Unfortunately, estimating the value of *A. sericeasur* suppression of the leaf rust and other pests would be difficult and require long term data due to the long term dynamics of the leaf rust that can result in severe yield losses for several years.

One common critique of the economic valuation of species is that they are overly simplified and their importance to the public and managers is therefore exaggerated. In our study, we attempted to improve upon some of these simplifying assumptions in our calculations. First, many assume

effect sizes produced by experiments or observational data can be extrapolated to the entire area of plantations, assuming that all foraging and effects are evenly distributed across plantations. This assumption is not valid in our study system, as our data show that large- and medium-sized colonies of *A. sericeasur* only occupy roughly 5% of trees in the 44 ha surveyed across 9 plantations. For other organisms with higher dispersal abilities, such as birds, there is some evidence that effects may be stronger near to forests (Karp et al. 2013). Valuations of the berry borer are based on a large assumption that berries bored are removed during processing and therefore a berry bored is a berry lost. However, many coffee farmers and processors sell different qualities of coffee, therefore it is more likely that bored coffee is not discarded but sold as lower quality coffee. Further, different coffee qualities sell for different values, which hugely impacts the estimation of pest control value. For that reason, we reported a range of values per pound of roasted or export quality coffee. Future studies should aim to evaluate the assumptions of species valuations and aim to provide valuations of pest control of the entire pest community, instead of focusing on individual pests.

Complex interaction cascades are surfacing across a plethora of systems. For example, McCauley et al. (2012) describe how the presence of native or exotic tree communities on islands have profound influences on the nutrient influxes into island ecosystems. Native tree communities have greater bird abundance, which leads to greater nutrient flux to the island, greater nutrient runoff into near-shore marine habitats, and increase phyto- and zooplankton abundance. The greater plankton abundances were correlated with the abundance of manta rays in proximity to natively vegetated islands. In addition, human settlement and agricultural practices are clear drivers of other interaction cascades. In Kenyan savannas for example, temporary fenced settlements, which protect livestock from predators at night, concentrate dung and result in long-term high-nutrient conditions at the site. This, in turn, drives an interaction cascade of greater tree size, greater arthropod abundance, and greater density of geckos (Donihue et al. 2013). Wielgoss et al. (2014) show how complex interactions between ants, a pest community, cacao pathogens, and cacao interact to impact cacao yield. High evenness in the ant community was correlated with the greatest marketable yield via a complex web of interaction pathways. Our study builds on the previous work of others (Vandermeer et al. 2010), and

together, these studies provide another example of complex interaction cascades in agricultural systems.

We also suggest that management for diverse tree communities with both *Inga* and non-*Inga* species may maximize control of multiple pests, while minimizing impacts of trees on coffee yield. If leaf rust and/or berry borer outbreaks are low, *Inga* trees may provide stronger production services. However, if the berry borer or the leaf rust reaches epidemic levels, increased numbers of *A. sericeasur* colonies nesting in non-*Inga* trees will provide coffee with greater protection. Further, reduction of shade tree canopy complexity, provided by tree species diversity, is associated with loss of bird, bat, and pollinator abundance and diversity (Perfecto et al. 2007, Jha and Vandermeer 2010, Williams-Guillen and Perfecto 2010). Thus, increasing tree canopy complexity may promote pollinators and birds that provide pollination and pest control services (Klein et al. 2003, Perfecto et al. 2004, Karp et al. 2013).

Complex interaction chains and cascades exist throughout ecosystems around the globe (Pringle et al. 2007, McCauley et al. 2012) and human activities disrupt and modify these interaction chains (McCauley et al. 2012, Donihue et al. 2013). Our study highlights that these long interaction chains are also important and context dependent in agricultural ecosystems and that the resulting services trade-off under different scenarios. Our work provides additional evidence that simplifying agricultural systems may result in unforeseen ecological and economic consequences (Howard 1940, Altieri 1987, Lewis et al. 1997, Tscharntke et al. 2005, Gliessman 2007, Power 2010, Vandermeer et al. 2010, Kremen et al. 2012) and that we may not necessarily know the importance of maintaining higher biodiversity levels given our limitations in quantifying the complexity of all species, their interactions, and the services they provide (Hector and Bagchi 2007; Isbell et al. 2011).

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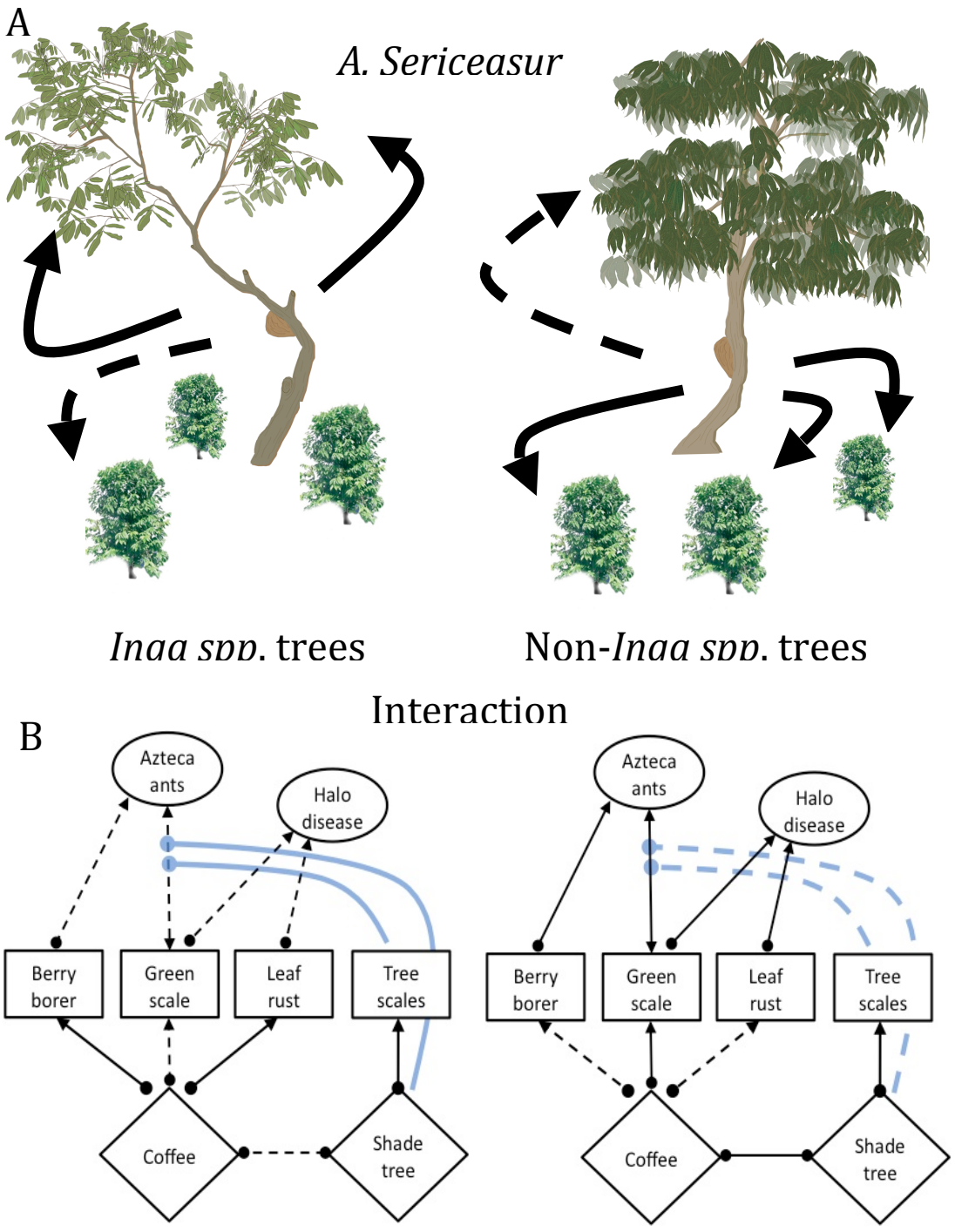

Figure 6.1. (A) The foraging direction of *A. sericeasur* in *Inga* and non-*Inga* trees and (B) the resulting interaction strengths between web components. Solid lines = strong interactions; dotted lines = weak interactions; arrows = positive effects, circles = negative effects, black lines = direct interactions, blue lines = indirect interactions. Trees designed by Z. Hajian-Forooshani.

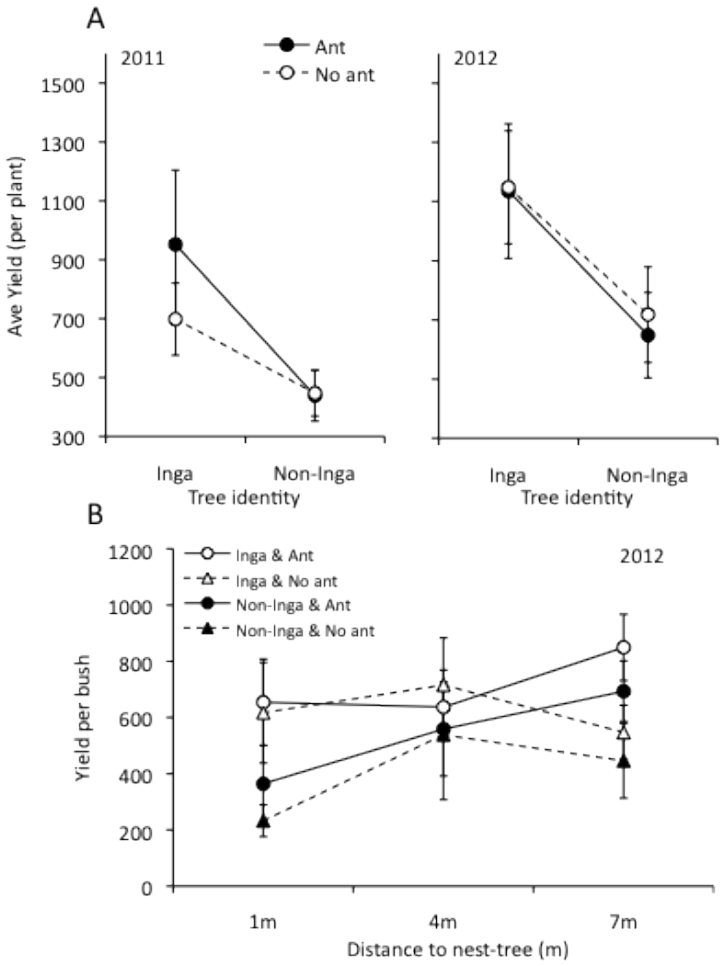

Figure 6.2. (A) Yield per coffee bush on *Inga* and non-*Inga* trees with and without *A. sericeasur* across 2011 and 2012. (B) An extra survey in 2012 of yield per coffee bush across *Inga* and non-*Inga* trees with and with *A. sericeasur* at 1-7m away from nest-trees.

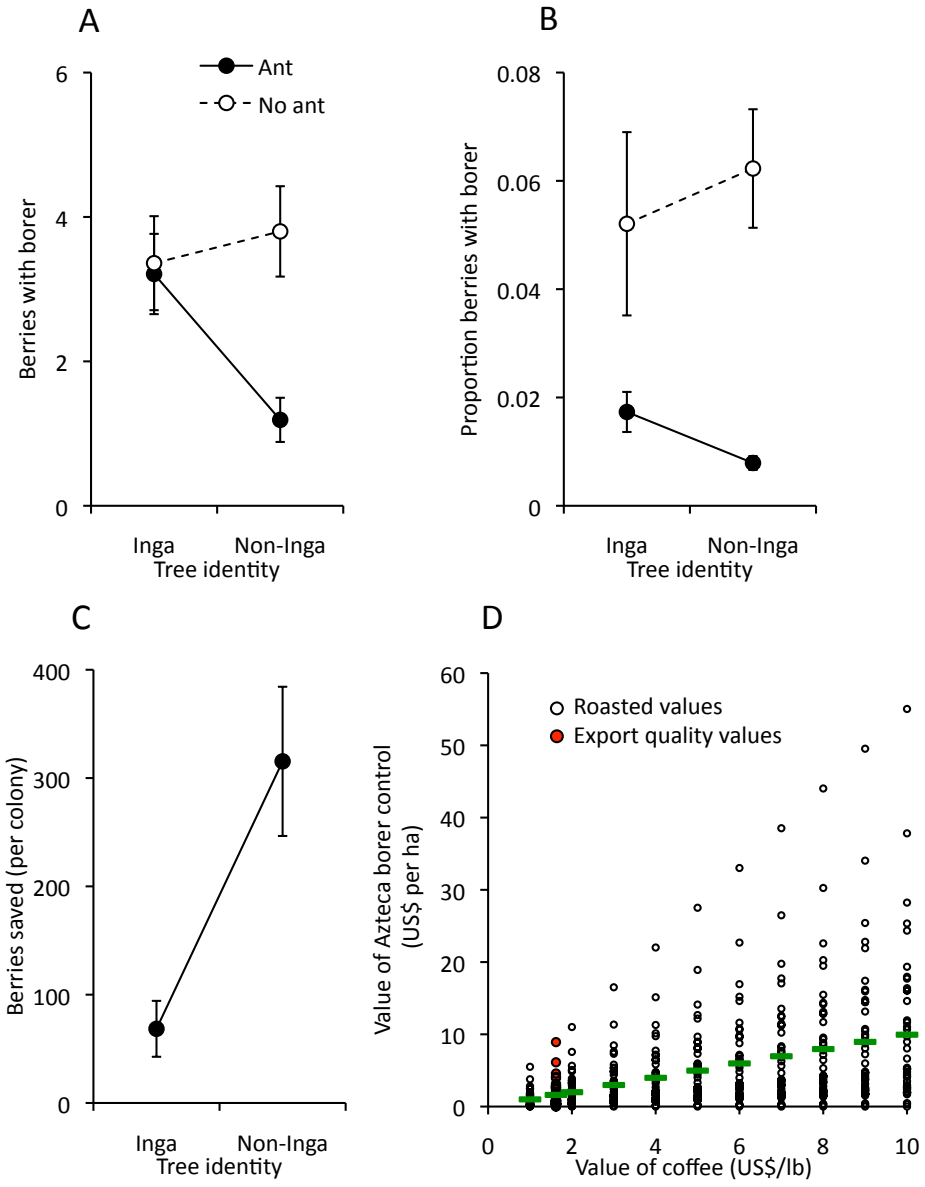

Figure 6.3. (A) Borer addition experiment comparing the number of berries with borers on coffee bushes with or without *A. sericeasur* colonies nesting in *Inga* or non-*Inga* spp. trees. (B) Two-year survey of the proportion of berries with borers on coffee bushes with or without *A. sericeasur* colonies nesting in *Inga* or non-*Inga* spp. trees. (C) Berries saved per colony = Δ borer × yield × bushes. Where Δ borer is the proportion of berries with borers in control (no ant) bushes subtracted by the proportion berries with borers in ant bushes (from two-year survey), yield is the average yield per bush for ant bushes, and bushes is the number of bushes foraged by the ant colony. All data is represented by mean ± standard error of the mean. (D) The estimated value of *A. sericeasur* pest control of the berry borer (US$ per ha) across 44 hectares surveyed and given different potential values of coffee sold.

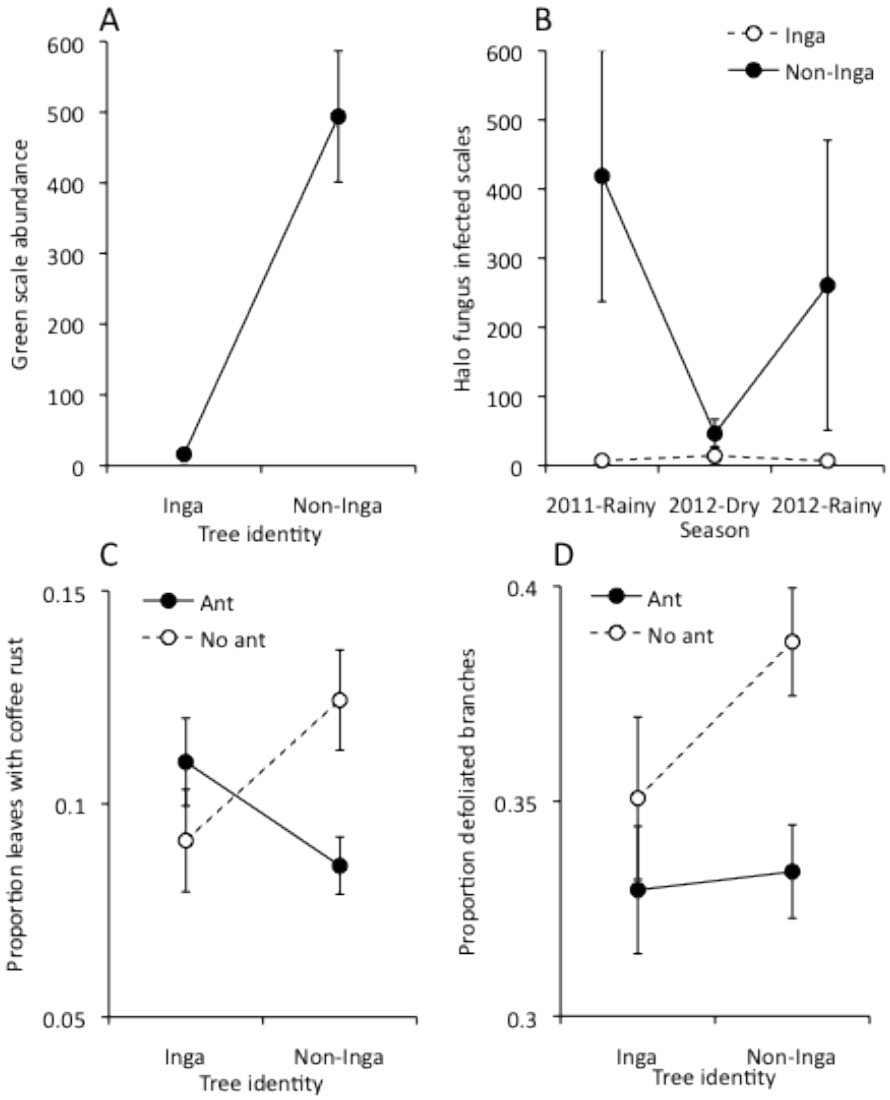

Figure 6.4. (A) Abundance of green scales (average 2011-12), (B) white halo fungus infected green scales (2011-12), (C) proportion of leaves with coffee rust (2013), and (D) the proportion of defoliated branches (2013) in coffee with *A. sericeasur* in *Inga* and non-*Inga* trees.

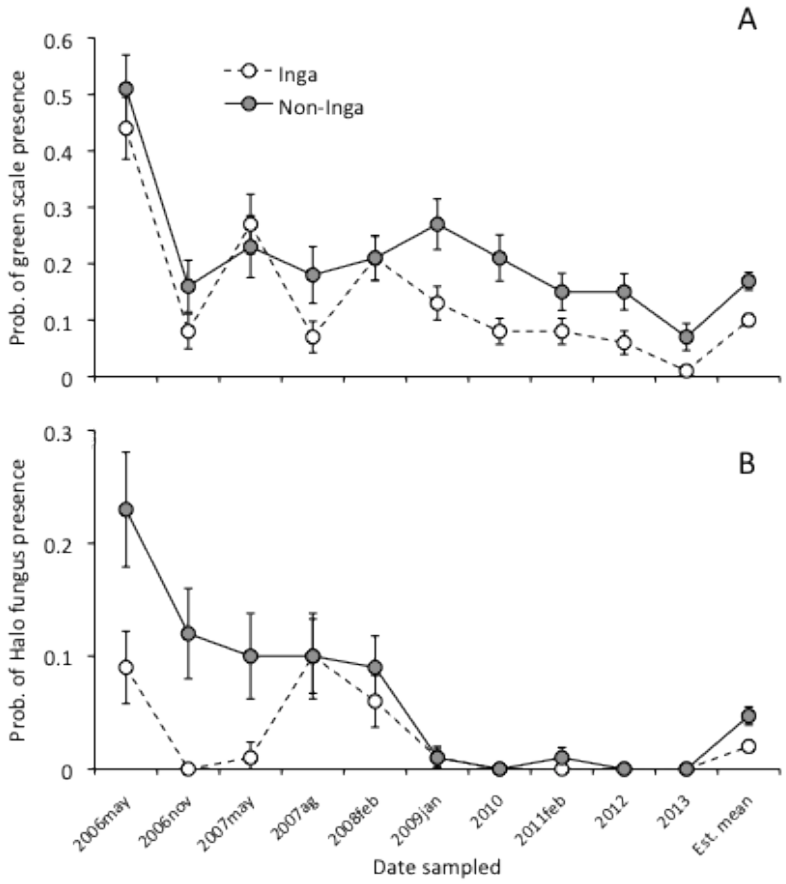

Figure 6.5. (A) The probability of *C. viridis* population occurrence and (B) the probability of white halo fungus occurrence in coffee plants around *A. sericeasur* colonies in *Inga*- and non-*Inga* spp. shade trees across 8 years of 45-ha plot surveys.

Table 6.1. Study outline and sample sizes.

		2011		2012				2013	
		Rainy		Dry		Rainy		Dry	
	A. sericeasur treatment	Inga	Non-Inga	Inga	Non-Inga	Inga	Non-Inga	Inga	Non-Inga
Detailed survey (4 plantations)									
Ant nest activity	*A. sericesaur*	15	16			16	17		
	no *A. sericeasur*	-	-			-	-		
Shade tree ant foraging	*A. sericesaur*	14	16	13	15				
	no *A. sericeasur*	-	-	-	-				
Shade tree honeydew producers	*A. sericesaur*	15	16	10	13	18	18		
	no *A. sericeasur*	-	-	-	-	-	-		
Berry borer addition experiment	*A. sericesaur*					15	15		
	no *A. sericeasur*					15	15		
Berry borer survey	*A. sericesaur*	15	16			16	17		
	no *A. sericeasur*	15	16			16	17		
Ant-borer pest control valuation	*A. sericesaur*	15	16			16	17		
	no *A. sericeasur*	15	16			16	17		
Coffee yield	*A. sericesaur*	15	16			16	17		
	no *A. sericeasur*	15	16			16	17		
Coffee green scale survey	*A. sericesaur*	15	15	13	14	15	17		
	no *A. sericeasur*	-	-	-	-	-	-		
White halo fungus survey	*A. sericesaur*	15	15	13	14	15	17		
	no *A. sericeasur*	-	-	-	-	-	-		
Leaf rust survey	*A. sericesaur*							12	13
	no *A. sericeasur*							7	10

Table 6.2. Summary of Ant colony, nest-tree, coffee, and site characteristics.

Tree type	*Inga*	Non-*Inga*	P
Tree connections	2±0.3	2.5±0.4	NS
Tree height	10±1	12±1	NS
Ant canopy activity (per min.)	**7.5±1.5**	**3.7±0.8**	*
Tree honeydew producers (per 1000 cm^2 leaf area)	**74±12**	**13±4**	**
Colony activity index	41±6	37±4	NS
Coffee density	33±2	36±2	NS
Coffee foraged (per bush)	**5.3±0.5**	**10.7±1**	***
Coffee ant activity (ants per min.)*	**4.4±0.7**	**4.7±0.5**	*

*Significant in 2012 rainy season, but not 2011.

Table 6.3. Full statistical models for shade tree, coffee and ant colony variables.

	$df_{1,2}$	F	P
Tree connections (Poisson)			
Tree type	1,93	0.1	0.767
Tree height (Gaussian)			
Season	2,91	3.3	**0.042**
Tree type	1,91	3.6	0.061
Season*type	2,91	0.1	0.875
Log canopy ant activity (Gaussian)			
Season	1,26	19.4	**<0.001**
Tree type	1,28	7.3	**0.012**
Season*type	1,26	0.4	0.529
Colony activity index (+)	1,27	7.2	**0.012**
Nest-tree honeydew producers (per 1000m cm^2) (Gamma)			
Season	2,78	9.4	**<0.001**
Tree type	1,78	10.3	**0.002**
Season*type	2,78	2	0.141
Coffee density (-)	1,78	1.2	0.277
Tree height (-)	1,78	3.3	0.075
Tree connections (-)	1,78	4.5	**0.036**
Colony activity index (+)	1,78	35.2	**<0.001**
Colony activity index (Gaussian)			
Season	1,21	18.4	**<0.001**
Tree type	1,37	0.3	0.562
Season*type	1,21	0.2	0.648
Coffee density (Gaussian)			
Tree type	1,43	2.4	0.132
Coffee bushes foraged (per nest) (Gaussian)			
Season	1,58	13.8	**<0.001**
Tree type	1,58	18.7	**<0.001**
Season*type	1,58	2.8	0.099
Colony activity index (-)	1,58	0.2	0.64
Coffee density (+)	1,58	5.1	**0.028**
Coffee ant activity (Gaussian)			
Season	1,37	8.4	**0.006**
Tree type	1,20	0.2	0.684
Season * type	1,32	7.2	**0.012**
Colony activity index (-)	1,43	<0.1	0.952
Distance to nest-tree (+)	1,37	1.3	0.262
Coffee density (-)	1,19	0.9	0.362

Table 6.4. Statistical analysis of yield per bush across season, tree type, and in the presence or absence of *A. sericeasur.* The second survey also monitored distance to nest-tree.

	df	F	P
2y survey data (yield per bush)			
Season	1,122	13.2	**<0.001**
Tree type	1,122	8.3	**0.005**
Ant presence	1,122	<0.1	0.942
Season*type	1,122	0.1	0.78
Season*ant	1,122	<0.1	0.896
Type*ant	1,122	0.8	0.377
Season*type*ant	1,122	0.5	0.473
2012 (extra yield data)			
Tree type	1,22	4.6	**0.043**
Ant presence	1,20	1.2	0.295
Distance to nest-tree	2,40	2.3	0.116
Type*ant	1,22	0.2	0.691
Type*distance	2,40	1.2	0.304
ant*distance	2,40	1.6	0.209
Type*ant*distance	2,40	0.1	0.909

Table 6.5. Effect of tree type and ant presence on berry borer infestation of coffee.

Borer addition experiment			
Berries with borers (Poisson) (2012)	$df_{1,2}$	F	P
Tree type	1,55	2.2	0.141
Ant presence	1,55	7.8	**0.007**
Type*ant	1,55	8.3	**0.006**
Borer survey 2011-12			
Prop. berries with borers (Binomial)		(Z)	
Season	1,86	-0.6	0.578
Tree type	1,86	-2	**0.044**
Ant presence	1,86	2.4	**0.015**
Type*ant	2,86	2.5	**0.012**
Season*ant	2,86	0.8	0.441
Season*type	2,86	0.1	0.928
Season*ant*type	3,86	-0.4	0.717
Colony pest control estimate			
Berries saved (per colony) (Gaussian)			
Season	1,57	0.6	0.46
Tree type	1,57	8.9	**0.004**
Season x type	1,57	<0.1	0.967
Colony activity index (-)	1,57	0.5	0.49
Coffee density (+)	1,57	4	**0.05**

Table 6.6. Statistical models for coffee green scales, the white halo fungus (*L. lecanii*), the proportion of coffee branch defoliation, and the proportion of leaves with the leaf rust.

	df	F	P
Coffee Scale density (Neg. Binomial)			
Season	2,79	2.5	0.088
Tree type	1,79	112.6	**<0.001**
Season*type	2,79	2.4	0.098
Colony activity index (+)	1,79	3.9	0.052
***L. lecanii* prevalence (Neg. Binomial)**			
Season	2,79	0.4	0.645
Tree type	1,79	23.1	**<0.001**
Colony activity index (+)	1,79	0.1	0.829
Season*type	2,79	3.4	**0.037**
Proportion branches defoliated (2013)		(Z-stat)	
Tree type	1,121	-2.0	**0.045**
Ant presence	1,121	2.8	**0.006**
Type*ant	1,121	-0.3	0.765
Proportion leaves with leaf rust (2013)		(Z-stat)	
Tree type	1,121	-9.0	**<0.001**
Ant presence	1,121	-3.2	**0.001**
Type*ant	1,121	7.3	**<0.001**

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Chapter 7
Conclusions

Agricultural ecosystems, like 'natural' ecosystems, are made up of a set of species interactions within ecological networks and the breakdown, modification or entire re-distribution of these interactions can have consequences for ecosystem functions and inevitably the value of that system to humanity. Some argue that complexity in agricultural systems is generally beneficial for aiding regulation of ecosystem services autonomously at desirable levels (Howard 1940, Altieri 1987, Lewis et al. 1997, Tscharntke et al. 2005, Gliessman 2007, Power 2010, Vandermeer et al. 2010, Kremen et al. 2012). This dissertation provides new insights into how changes to agricultural management impact the diversity of species (Chapter 2) and trophic dietary niche of species (Chapter 3). It also provides analysis of how multiple species perform pest control services (Chapter 4 & 5) and how context dependency in interaction webs can drive cascading effects on pest control and other services (Chapter 6).

Chapter 2 provides general insight into how agriculture impacts diversity in different taxonomic groups. It suggests that larger, more mobile taxonomic groups like birds are impacted by the larger spatial-scale impacts of agriculture, such as the homogenization of landscapes (greater percentage of agricultural land-uses) or reduced diversity of habitats in the landscape. It also shows that sessile groups, like plants are strongly influenced by local level management factors. These findings are important for understanding how to better manage agricultural landscapes to maintain species for the value of the services they provide or for the sake of conserving the abundance and richness of species in these heavily human-impacted habitats.

Chapter 3 provides a more in-depth look at how species are affected by management changes and may help us understand the connections between the loss of resources and the local extinction of species following the simplification of agroecosystems. It shows that the trophic

niche width of four ant species was constricted following simplification of coffee agroecosystems. It also shows that one species shifted trophic position and that there was a change in the overlap in trophic position between species. Given the importance of competition in ant ecology (Hölldobler and Wilson 1990) these results suggest changes in niche overlap may be one driver of community change following intensification. The results also suggest that a species' ecological function within the agroecosystem may change even if the abundance of a given species is unchanged by intensification. This could lead to greater or weaker functioning and provisioning of ecosystem services for those species. These results also corroborate the findings of Edwards et al. (2013) who found degradation of forests from old-growth to secondary-forest limited the trophic niche width of bird species and species with the narrowest niche widths were limited to only the old-growth habitats. Agricultural intensification, like forest degradation, is hypothesized to limit the diversity and abundance of species through reducing the diversity and availability of resources. Thus, the fact that all ant species studied had lower trophic niche widths in intensified habitats provides some of the best data, to date, to support this hypothesis.

Chapter 4 provides insights into the importance of considering all potential pest control agents in agroecosystems. Few consider ants as major pest control agents, although historically they have been important players (Van Mele 2008). This study provides evidence that in coffee production systems 6 of 8 ant species limit the colonization of coffee berries by the berry borer. The results of this study emphasize that managers should consider many potential pest control agents and not only focus on specialized predators or parasitoids of pests.

Chapter 5 provides empirical support of theoretical hypotheses (Wilby and Thomas 2002, Briggs and Borer 2005) that suggest diverse communities of consumers are more efficient at extracting diverse resource pools than are simple consumer communities. Increased resource diversity should promote the ability of multiple predator species to partition prey-resource-niches driving greater effectiveness of diverse compared to simple predator communities, however it is very difficult to test with predator species. Our study shows that multiple predators were only more efficient at controlling pest communities relative to single predator species, when pest communities were diverse. This result has major implications for the framing of how pest control

research is undertaken. The majority of multiple predator experiments in natural and agricultural ecosystems study predator effects on single prey species, even though most prey communities are made up of many more prey species. Of the multiple predator experiments in agricultural systems that were reported in a recent review (Cardinale et al. 2006, Griffin et al. 2013), 66% (39/59) of studies focus on one prey (herbivore) species and just one experiment included more than two prey species. Further, 93% of experiments focus on aphid species only, while the remaining four investigate leafhoppers or planthoppers (other families within the Hemiptera order), suggesting substantial taxonomic bias (Cardinale et al. 2006, Griffin et al. 2013). While many contend that it is important to focus on major pests to understand how to limit economic damage, it is also arguable that the importance of each pest species waxes and wanes because pest populations are dynamically changing over time and space (Lewis et al. 1997). Thus the goal of predator diversity experiments should be to evaluate the importance of predator communities at regulating the entire pest communities. Further, our results reflect that if more diverse pest communities are considered then more diverse predator communities are needed to control this diversity.

Chapter 6 reveals how the strength of interaction cascades through complex-interaction-webs are context dependent and how different scenarios promote different suits of services. Complex interaction chains and cascades are being uncovered in many ecosystems around the globe (Pringle et al. 2007, McCauley et al. 2012). Other studies have highlighted how human activities are disrupting or modifying these interaction chains (McCauley et al. 2012, Donihue et al. 2013). Like a similar study in cacao plantations of Indonesia (Wielgoss et al. 2014), our study shows that indirect effects driven by ants in agricultural systems are important in providing services and disservices. Our study also reveals how trade-offs between multiple services are the result of the management of shade tree species. It highlights the importance of considering non-trophic effects in interaction-webs, as the documented context-dependent effects appear to be driven by ant-hemipteran mutualisms in the shade tree canopies and on coffee. Like other recent studies, it also provides evidence to suggest that ant-plant associations are important to plant pathology in both direct and indirect and negative and positive pathways (Vandermeer et al. 2010, González-Teuber et al. 2014, Vandermeer et al. 2014, Wielgoss et al. 2014). The trade-offs between shade-tree services and ant pest control services are extremely important to the literature that aims to

understand the multifunctionality of land-use, agricultural management, and biodiversity (Foley et al. 2005, Tscharntke et al. 2011, Iverson et al., in prep.)

Synthesis

This dissertation provides several examples of why management in agriculture can influence the number of species (Chapter 2) and the strength of interactions that occur within ecological communities, and provides several examples of why ecological complexity matters in agriculture (Chapters 3-6). Focusing solely on therapeutic methods of controlling the most damaging pests in agriculture has often resulted in the promotion of chemical and biological control practices that are aimed at eradicating major pests. These practices often ignore or even reduce the effectiveness of other biological control agents and/or are so specific that other pest species can rise to become problematic (Lewis et al. 1997). Instead, many have argued that it is more important to understand the ecological system and why certain management practices result in pest outbreaks and why others lack damaging levels of pest species (Howard 1940, Altieri 1987, Morales et al. 2001). A number of ecological studies are now being presented that suggest that managing for diversity or high evenness within pest control communities promotes autonomous control of the entire pest community by regulating populations at lower densities that are not damaging to crops (Crowder et al. 2010, Vandermeer et al. 2010, Wielgoss et al. 2014). This dissertation joins these studies to provide further examples of why it is important to consider diversity and complexity in agriculture. This work suggests that non-trophic interactions (ant-hemipteran mutualisms) are important and that minor pests provide resources to predators that limit more damaging pests. Further, this dissertation finds that diverse predator communities are better at suppressing multiple pests simultaneously compared to individual predator species, suggesting that in real agricultural ecosystems where typically there are many pest species, management for multiple predator species will be important. Thus, this dissertation finds that diversity and ecological complexity is important to the provisioning and regulating services delivered to agriculture, but also warns that non-trophic and cascading effects may not always be predictable, as has been reflected by others (Berlow et al. 2009). Additionally, this dissertation suggests it is important to consider multiple ecosystem services within systems, as there are often trade-offs between services (Power 2010, Letourneau et al. 2011, Iverson et al., in prep.).

Documenting these trade-offs can provide greater insight into to how to better manage for all services in systems with different pressures.

As in other fields, researchers in agriculture have long considered the importance of ecological complexity (Howard 1940, Altieri 1987, Lewis et al. 1997, Gliessman 2007, Vandermeer 2010). However, the mechanization and industrialization of agricultural production systems have increased yields at the expense of ecological complexity within fields and farming landscapes. While many marvel at modern agriculture's incredible efficiency and high yields, these modern productions systems introduce many agricultural and environmental problems. Given that even intensive farms are ecosystems, ecological theory and practice may help provide solutions to the associated problems. This dissertation provides insights into how ecological complexity is important to consider in the agricultural production of the tropical crop coffee.

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