

EVALUATION OF NATURAL ENEMIES IN BLUEBERRY AGROECOSYSTEMS:  
IMPLICATIONS OF MANAGEMENT PRACTICE AND SURROUNDING  
LANDSCAPE

by

TYLER SETH WHITEHOUSE

(Under the Direction of Jason M. Schmidt)

ABSTRACT

Natural enemies are valuable components of agroecosystems as they provide biological control services to help regulate pest populations. Promoting biocontrol services can improve sustainability by decreasing pesticide usage, which is a major challenge for the blueberry industry in the southeastern United States. We conducted this study across the most productive blueberry producing counties in Georgia, USA during the growing seasons of 2016 and 2017. Blueberry orchard sites were selected to compare multiple factors including management practice, local vegetation, and landscape diversity. Natural enemies were found to be more abundant in the surrounding landscapes and vegetation between crop rows supported higher natural enemy populations. Our results suggest implementing ecologically based management practices in order to sustain diverse natural enemy communities and potentially limit pest populations, including spotted wing drosophila (*Drosophila suzukii*).

INDEX WORDS: Araneae, biological control, blueberry, *Drosophila suzukii*, ecosystem services, parasitoids, predator distribution, spiders

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TYLER SETH WHITEHOUSE

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TYLER SETH WHITEHOUSE

Major Professor:	Jason M. Schmidt
Committee:	Ashfaq A. Sial
	Kerry M. Oliver

Electronic Version Approved:

Suzanne Barbour  
Dean of the Graduate School  
The University of Georgia  
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## DEDICATION

I would like to dedicate this work to my dearest friends and family, whose support and motivation have helped me to pursue my passion and spread my wings.

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## CHAPTER 1

### INTRODUCTION AND LITERATURE REVIEW

Blueberry production in the southeastern region is a relatively new commodity that has become widely successful and has continued to expand. Like all growing agricultural systems, agricultural intensification comes with adverse effects for the fauna of the region. Considerations must be made towards improving sustainable management and providing suitable habitat within agroecosystems. This research will be the first to evaluate the natural enemy community within blueberry orchards of the southeast United States. The goal of this research is to evaluate the effects of local and landscape factors on natural enemy abundance and distribution, while also assessing how the major pest of concern is impacted. The proposed research will facilitate growers in the adoption of integrated pest management strategies towards improved biocontrol services provided by natural enemies.

#### **Local factors**

The first local factor evaluated is the chemical management practice, in which blueberries across the nation are produced in either conventional practices or organic practices. Conventional practices are mostly comprised of large scale systems with high intensity use of synthetic, broad-spectrum chemicals (Boutin et al. 2008). Organic practices are limited to reduced-risk insecticides, which have been shown to favor natural enemies when compared to conventional practices in other agricultural cropping systems (Boutin and Jobin 1998, Sciarappa et al. 2008, Koss et al. 2005, Crowder et al. 2010,

Cardenas et al. 2015, Martin 2016). A study by Schroter and Irmeler (2013) examined how intensively managed fields prevent predators from entering the crop field, and the conversion to organic fields allowed predators to disperse more easily. Bioassay studies have also demonstrated the lethal effects of insecticides used in blueberry systems on natural enemies in the laboratory, however there is a need for supporting field trial data (Roubos et al. 2014). Unmanaged systems are unique to this study in that they act as a control, as they are essentially abandoned blueberry orchards without exposure to pesticides. There are few studies that have compared the effects on natural enemies between unmanaged systems to managed agricultural systems (conventional and organic). Grape vineyards, for instance, have shown higher abundances of natural enemies in unmanaged as compared to managed systems (Prischmann et al. 2005). The unmanaged vineyard contained fewer pest pressures with a higher diversity of natural enemies, which may be due to biocontrol services or simply fewer grapes for pests to utilize as hosts. The natural enemies in managed systems were most likely limited by the availability of resources and insecticide usage. Information regarding management practices will provide insight to how natural enemy populations are affected at the local scale, while suggesting the need for habitat management techniques.

Another local effect is habitat management, which includes the within field vegetation between rows, vegetation composition, and microclimate conditions (Landis et al. 2000 and Horvath et al. 2015). In the current blueberry system, many of the orchards suppress weeds or vegetation between crop rows through herbicide applications or frequent mowing. The application of herbicides causes indirect negative effects to natural enemies by removal of available resources and vegetation (Evans et al. 2010). The

benefit of vegetation has been examined in many different systems through different ground cover amendments between crops rows, thus proving bare ground to have negative effects on natural enemies (Mathews et al. 2004, Minarro et al. 2009, Silva et al. 2010, Cox et al. 2014, Fox et al. 2015). In particular, blueberry systems that compare within field management have only focused on ground-dwelling predators (O'neal et al. 2005, Renkema et al. 2012, Jones et al. 2016). Rather, our focus is on the natural enemies found in the canopy that depend on alternative resources within fields where refuge is required during chemical management and disturbances (Thomas and Marshall 1999, Thorbek and Bilde 2004, Thomson and Hoffmann 2009). Non-crop habitats can provide refuge and resources in response to very limited resources available in intensively managed crop fields. Habitat management focusing on spatial effects have mainly been studied in annual crops, where perennial agroecosystems are less examined. By manipulating the habitat to mimic natural, unmanaged practices with presence of vegetation between crop rows, natural enemies may be maintained in the crop area (Landis et al. 2005, Isaacs et al. 2009).

### **Landscape factors**

A term quite frequently used in Europe is agri-environment schemes, which is a method of conserving biodiversity in farmland (Fahrig et al. 2011). In order to implement this landscape management tactic, the landscape factors often examined are compositional heterogeneity (proportions of different habitat types) and configurational heterogeneity (arrangement of habitats). Landscape composition has been shown to be the most important in shaping natural enemy populations as they provide overwintering and refuge from human disturbances (Clough et al. 2005 and Schmidt et al. 2005). As the

surrounding landscape is simplified, predator-prey interactions are destabilized, whereas habitat complexity allows managed systems to be continuously recolonized by natural enemies throughout the growing season (Landis 2016). Interestingly, many studies have shown that the relative importance of landscape factors depends on the mobility of the taxa examined (Perovic et al. 2010 and Gonthier et al. 2014). This raises the importance of determining which environmental factors play the largest role in their respective ecosystem. It is hypothesized that more complex habitat types at the landscape level and more suitable habitat at the local level couple to provide more biological control services in the agroecosystems (Veres et al. 2013, Aviron et al. 2016, Bianchi et al. 2006, Tscharrntke et al. 2005, Chaplin-Kramer et al. 2011). Many studies analyze natural enemies without considering the alterations of the primary pest of concern, which is necessary to calculate the tradeoffs of incorporating new management tools.

### **Blueberry crop and associated pests**

Blueberry production in the southeastern U.S. is mostly made up of southern highbush (*Vaccinium corymbosum*) and rabbiteye (*Vaccinium ashei*). These blueberry species have allowed Georgia to become one of the most successful producers in the United States and has shown consistent growth from 59 million lbs in 2011 to 96 million lbs in 2014 (Melancon 2014). Over 20,000 acres are used for production in Georgia, which presents a variety of challenges for pest management (NASS 2014). The common blueberry pests include the blueberry gall midge (*Dasineura oxycoccana*), glassy-winged sharpshooter (*Homalodisca vitripennis*), blueberry maggot (*Rhagoletis mendax*), and various thrips species, all of which have caused considerable damage to blueberries in the southeastern U.S. (Turner and Liburd 2007, UGA Extension 2017). The array of pests is

capable of damaging blueberry crops at different stages. For example, *D. oxycoccana*, *H. vitripennis*, and thrips occur during the pre-bloom or bloom stage and *R. mendax* occurs during pre-harvest before the production, harvest period (UGA Extension 2017). *D. oxycoccana* and thrips destroy flower buds and vegetative growth, *H. vitripennis* transmits bacterial leaf scorch disease, and *R. mendax* infests berries. However, more recently an invasive species, spotted wing drosophila (*Drosophila suzukii*), has raised the most concern. *D. suzukii* has quickly spread and infested fruit crops across North America since its first detection in 2008 (Asplen et al. 2015). This invasive species is able to deposit its eggs into ripening fruit causing a dramatic decrease in yield due to the fruit market's zero tolerance policy of infested fruit. The first and most vital management step is monitoring (Grant and Sial et al. 2016). There are many monitoring strategies and trap designs available to detect presence of *D. suzukii* including synthetic lures, yeast and sugar mixtures, and apple cider vinegar. The yeast and sugar bait trap is the most widely used and considered to be the most favorable monitoring method for *D. suzukii* (Landolt et al. 2012). Once *D. suzukii* is detected within a blueberry orchard, the first option is often chemical control. The most effective insecticides are broad-spectrum insecticides used in conventional practices (Van Timmeren and Isaacs 2013). Unfortunately, insecticides only cause lethal effects for adult *D. suzukii* and application rates have increased by up to 50% since its first detection, as larvae within the berry may survive various applications with nearly 13 generations year round in the southeastern U.S. (Diepenbrock et al. 2016). As more insecticides are applied and blueberry production increases, more information is needed regarding *D. suzukii* distribution. *D. suzukii* is capable of utilizing native fruit crops (Haviland et al. 2016 and Klick et al. 2016), thus

allowing *D. suzukii* to migrate between landscapes with an abundance of host resources in the environment (Grant and Sial 2016). Knowledge of *D. suzukii* preference of different land covers and distribution throughout the agroecosystem can be incorporated into integrated pest management tactics.

### **Biological control**

When natural enemy populations are disturbed, it ultimately leaves regions susceptible to pest outbreaks. In order to effectively combat pest encroachment in a growing blueberry industry, sustainable approaches are needed to utilize biocontrol services. Natural enemies have been found to dramatically reduce pesticide usage, thus greatly contributing to integrated pest management systems (Tscharntke et al. 2007). Habitat management is an important tool in promoting natural enemies, for example the presence of vegetation surrounding and within the crop field can maintain predator populations (Fiedler et al. 2008, Walton and Isaacs 2011, Gareau et al. 2013, Blaauw and Isaacs 2015). In particular, we know that ground-dwelling predators in blueberry systems are impacted by habitat management including bare ground, mulch, and vegetation (O'neal et al. 2005, Renkema et al. 2012, Jones et al. 2016). The information regarding these impacts are vital to providing biological control programs through the implementation of habitat management. As agricultural intensification continues to dictate the natural enemy populations, this allows pests to more easily invade within the crop fields (Hogg and Daane 2010, Landis et al. 2005). Generalist predators, such as spiders, contribute greatly to biological control of pests in many agroecosystems (Symondson et al. 2002, Richardson and Hanks 2009, Cardenas et al. 2015, Picchi et al. 2016). Spiders are often the dominant predators in many agroecosystems and have been

shown to limit pests in perennial systems. Different functional roles are played by different groups of spiders where active hunters and web-building taxa may consume a variety of pests (Nyffeler 1999). However, this important group of predators is heavily influenced by insecticides and availability of suitable habitat (Picchi et al. 2016). To combat the negative impacts of management, non-cropping native plants are shown to promote spider communities and pest control potential (Amaral et al. 2016). Initial data on natural enemy populations in blueberry system will provide the foundation of knowledge for understanding and developing biocontrol options.

Another method of biocontrol required for this region is parasitoid activity. Parasitoids of the most common blueberry pest, *D. suzukii*, have been discovered in other regions of North America, yet there have been no direct observations made in the southeastern United States. The most likely Hymenoptera parasitoids are those in the genus *Ganaspis*, *Leptopilina*, *Trichopria*, and *Asobara* that are found to be natural enemies of *D. suzukii* in their native range (Mitsui et al., 2007). There have been few reports in North America where two generalist parasitoids, *Pachycrepoideus vindemiae* and *Trichopria drosophilae*, successfully emerge from *D. suzukii*. The parasitoids of the genus *Trichopria* lay eggs directly into the hemocoel of spotted wing drosophila pupae and ectoparasitoids of the genus *Pachycrepoideus* lay eggs in the pupal case of spotted wing drosophila (Carton et al. 1986). These parasitoids are able to effectively parasitize *D. suzukii* located within the infested fruit and often compete with each other for host usage (Wang, Xin-Geng, et al., 2016). This project has also prepared the collected parasitoids towards future analysis for identification purposes.



## **Project objectives and approach overview**

- 1. Evaluate how different management practices affect natural enemy abundance and distribution.**
  - a. Select blueberry orchards and sample for natural enemies along a transect located within each orchard to test for distribution of natural enemies.
  - b. Determine the abundance of natural enemies among three management practices (conventional, organic, and unmanaged).
  - c. Determine the temporal and spatial distribution on natural enemies related to management practices.
- 2. Assess the effects of habitat management and the surrounding landscape on natural enemies and spotted wing drosophila (*Drosophila suzukii*).**
  - a. Select blueberry orchards to represent a range of landscape characteristics.
  - b. Determine the effects of local factors within the orchard and the impacts on arthropod abundance and distribution.
  - c. Measure landscape factors through geospatial tools to quantify the surrounding landscape and indicate the relative importance of each factor on the abundance of natural enemies and *D. suzukii*.

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CHAPTER 2

NATURAL ENEMY ABUNDANCE IN SOUTHEASTERN BLUEBERRY  
AGROECOSYSTEMS: DISTANCE TO EDGE AND IMPACT OF MANAGEMENT  
PRACTICE

**Abstract**

Natural enemies are valuable components of agroecosystems as they provide biological control services to help regulate pest populations. Promoting biocontrol services can improve sustainability by decreasing pesticide usage, which is a major challenge for the blueberry industry. Our research is the first to compare natural enemy populations in managed (conventional and organic) and unmanaged blueberry systems, in addition to the effects of non-crop habitat. We conducted our study in 10 blueberry orchards during the growing season across the major blueberry producing counties in Georgia, U.S.A. To estimate the spatial distribution of natural enemies, we conducted suction sampling at three locations in each orchard: within the forested border, along the edge of blueberry orchard adjacent to forested border, and within the interior of the blueberry orchard. Natural enemies maintained higher abundance over the season in unmanaged areas as compared to organic or conventional production systems. In the conventional orchards, natural enemies were more abundant in the surrounding non-crop area compared to the interior of the orchard. Populations were more evenly distributed in less intensive systems (organic and unmanaged). Our results indicate spatial structure in natural enemy populations is related to management practice, and less intensive management can retain higher abundance of natural enemies in blueberry systems. Considerations must be made towards promoting ecologically based management practices to sustain natural enemy populations and potentially increase the delivery of biological control services.

**Key Words** Araneae, ecosystem services, parasitoids, predator distribution, spiders

## Introduction

Blueberries, *Vaccinium* spp., are an important commodity in North America, and production continues to increase (Strik and Yarborough 2005). The state of Georgia has recently become one of the top producers and has shown consistent growth from 59 million lbs in 2011 to 96 million lbs in 2014 (Melancon 2014). Increased production and acreage presents a variety of challenges for pest management as many larval insects contaminate the fruit prior to harvesting. A primary pest of concern is the spotted wing drosophila (*Drosophila suzukii*), which has quickly spread and infested fruit crops across North America (Asplen et al. 2015). Insecticide application rates have increased by 50% since the first detection of *D. suzukii* (Diepenbrock et al. 2016). In order to effectively combat pest encroachment in a growing blueberry industry, sustainable approaches are needed to conserve and promote biocontrol services. Natural enemies can dramatically reduce pesticide usage, contributing to reduced input costs and environmental risk (Tscharrntke et al. 2007). However, the vast majority of natural enemy surveys in blueberry orchards focus on ground-dwelling predators and were not conducted in the southeastern region. In order to improve biological control programs and sustainable pest management, knowledge of natural enemy community composition is needed for this major blueberry producing region.

Currently, growers depend solely on the use of insecticides to control pest populations, which disrupts integrated pest management (IPM) strategies and reduces the ability for natural enemies to effectively limit pests (Johnson 2009). Blueberries are produced using conventional practices, which include intensive use of synthetic, broad-spectrum pesticides (Boutin et al. 2008), or organic practices, which employ reduced-risk

insecticides (Sciarappa et al. 2008). Organic management practices promote natural enemy diversity compared to conventional practices (Weibull et al. 2003, Crowder et al. 2010, Cardenas et al. 2015), where broad-spectrum insecticides often cause higher mortality on natural enemies than pests (Cardenas et al. 2015). Minimally managed and abandoned orchards are unique perennial systems that can provide insight on how production practices, such as pesticide use, effect natural enemy populations. (Prischmann et al. 2005 and Horvath et al. 2015). To our knowledge, previous studies have not examined “unmanaged” blueberry orchards. However, other agriculture production systems, such as olive orchards and forage crops, show higher natural enemy abundance in unmanaged compared to managed systems (Prischmann et al. 2005 and Horvath et al. 2015), likely a function of soil and chemical disturbances associated with management practices.

Habitat management through the addition of natural vegetation and non-crop flowering plants is a tool used to recruit healthy populations of natural enemies into managed, cropping systems (Fiedler et al. 2008, Walton and Isaacs 2011, Gareau et al. 2013, Blaauw and Isaacs 2015). Non-crop vegetation within and surrounding agroecosystems play a large role in the spatial structure of natural enemy communities where immigration depends on the availability of resources (Schellhorn et al. 2014). Alternative food resources and refuge from insecticide applications may be situated along forested borders, thus driving higher abundances of natural enemies towards the edge of the agroecosystem. Grower adoption of non-crop habitat can enhance natural enemies (Landis et al. 2005, Isaacs et al. 2009, Fox et al. 2015) and potentially allow predators to forage throughout the agroecosystem from edge to interior. For instance, ground-dwelling

predators are more abundant throughout the blueberry system when more resources, such as mulch, are provided between blueberry rows (O'neal et al. 2005, Renkema et al. 2012, Jones et al. 2016).

Despite large-scale growth and importance of the blueberry industry in the southeastern U.S., information of the natural enemies present is lacking. The goal of our study was to characterize the natural enemy community structure, and to evaluate how different management practices shape abundance and distribution throughout the agroecosystem. Our approach compared managed to unmanaged systems which is vital to understanding factors that contribute to supporting natural enemies without human disturbance, and may foster strategies to improve delivery of biocontrol services in managed systems. We hypothesize that natural enemy abundance decreases as management intensity increases. We also expect within intensive management systems, predators are less impacted towards the forested border. Our study objectives were to: (1) characterize natural enemies found within southeastern blueberry agroecosystems, (2) determine the impacts of management practice on natural enemy abundance, and (3) evaluate the spatial distribution of natural enemies related to management practice.

## **Materials and Methods**

**Study Area.** We located 10 commercial blueberry orchards distributed in five counties across South Georgia, U.S.A. (Fig. 2.1). All counties are situated along the Satilla River Estuary and our study area included a spatial extent covering approximately 5,645 km<sup>2</sup>. The selected blueberry orchards consisted of *Vaccinium ashei* (rabbiteye blueberry) and *Vaccinium corymbosum* (southern highbush blueberry). Orchards ranged in size between

0.52 ha and 76.5 ha, which we calculated using ArcGIS Release 10.3.1. We compared three different management systems among blueberry orchards: conventional (4), certified organic (3), and unmanaged (3) (Fig. 2.1). Management classifications were made based on intensity and type of insecticides applied. The four conventional sites utilized broad-spectrum synthetic insecticides including primarily organophosphates and pyrethroids, and in some cases a spinosyn (i.e. delegate) with herbicides applied between the blueberry rows for weed management. The three organic sites utilized reduced-risk organically certified (OMRI listed) insecticides and mowed vegetation between blueberry rows, but did not apply herbicides. The three unmanaged sites did not utilize pesticides and varied from abandoned orchards to small scale harvesting with vegetation present between orchard rows mowed infrequently. Non-crop habitats surrounding sites consisted mostly of coniferous forests with dense shrubbery and blackberry (*Rubus* spp). The presence of vegetation between blueberry crop rows and herbicide application was determined by visual observation and discussions with each grower at all ten sites.

**Sampling.** Each orchard was sampled weekly along a transect containing three stations for a total of 30 samples per week. Transects include locations of 25 m into the crop (orchard interior), along the orchard margin or first crop row (orchard border), and 15 m within the bordering, non-crop forested habitat (forest). Each sample consisted of 30 seconds of suction sampling with a modified reversed-flow leaf blower (SH 86 C-E; Stihl, Waiblingen, Germany) containing a mesh bag over the intake to collect natural enemy communities in the canopy of the blueberry orchards. One entire blueberry plant was suction sampled at each location, and one non-crop block of 2 m<sup>2</sup> was suction

sampled in the adjacent forest border. The samples were transferred to plastic bags and placed on ice until permanent storage at  $-20^{\circ}\text{C}$  in the laboratory. Individual specimens were separated from samples, identified to taxonomical level (family or order), and preserved in 95% ethanol at  $-20^{\circ}\text{C}$ .

**Analysis.** To characterize the structure of predator communities found in and around blueberry orchards, natural enemy counts were grouped by feeding habits and taxonomically as either Arachnids, insect predators, or parasitoids to compare the impact of management practices on general patterns of natural enemy abundance. Most arachnids were identified to family with the exception of immature spiders lacking key features. All insect predators were identified to family, and parasitoids were grouped as parasitic Apocrita. Prior to formal analysis of overall natural enemy abundance patterns, we tested for spatial autocorrelation between mean overall natural enemy counts and commercial orchard sites using Mantel's test with coordinates specified as latitude and longitude for each field location (package = "ADE4"; R Core Team 2015). Analysis of natural enemy population variability (total natural enemy abundance) in blueberry agroecosystems was analyzed using Generalized Least Squares (glS) fitted with REML to account for repeated measures of communities within orchards overtime (i.e. random error partitioned with orchard site as random variable; i.e. 1| orchard site) (Pinheiro and Bates 2000). The response variable, total natural enemy abundance, was natural logarithm transformed prior to the analysis. The best fitting error structure that produced spread of residuals was "weights=varPower()". The analyses were performed using R



version 3.3.1 (R Core Team 2016) and significant main effects, management practice and transect, were analyzed with linear contrasts for multiple comparisons of mean responses.

## Results

We collected a total of 1,113 natural enemies from suction sampling, including: 857 arachnids, 128 insect predators, and 128 parasitoids (Table 2.1). Arachnids made up of Araneae, the true spiders, combined with Opiliones, harvestmen, were the numerically dominant predatory arthropod taxa with twelve families. Insect predators were composed of nine families and Hymenopteran parasitoids were made up of parasitic Apocrita (Table 2.1). The distribution of Arachnids as the dominate predator taxa was consistent across all three management practices (Table 2.1; Fig. 2.2).

Spatial autocorrelation was not found among our blueberry orchards for the abundance of natural enemies ( $r^2=0.005$ ,  $P=0.4373$ ). A combination of sampling date, management practice and orchard sampling location (transect) contribute to explaining variability of natural enemy populations in blueberry orchards (Table 2.2). Although orchard size ranged from 0.52 ha to 76.5 ha, transects among all sites remained the same size and differences in orchard size did not significantly correlate with natural enemy populations (Table 2.2). No significant interaction between orchard size and management practice indicated that any effects of orchard size were independent of management practice (Table 2.2).

Natural enemies accumulated over time in orchards were dependent upon management practice (Fig. 2.3). The beginning of the sampling period contained an average of 2 ( $\pm 0.88$ ), 2.67 ( $\pm 0.60$ ), and 4.1 ( $\pm 1.12$ ) predators per sample in conventional,

organic, and unmanaged, respectively. A significant interaction between week and management practice (Table 2.2) is explained by natural enemies increasing over the season in the organic (slope est.=0.12 (0.05), t-value = 2.16, p-value=0.03) and the unmanaged systems (slope est.=0.13 (0.05), t-value = 2.39, p-value=0.02), as compared to conventional where populations remained low the entire season (Fig. 2.3).

Management practice and within orchard transects significantly influenced natural enemy populations (Table 2.2; Fig. 2.4). Unmanaged blueberry sites contained the highest mean natural enemy populations 9.92 ( $\pm 0.94$ ) followed by organic 5.16 ( $\pm 0.71$ ), and conventional 2.14 ( $\pm 0.26$ ). Orchard transects within management practices also showed significant differences in the natural enemy community. In conventional orchards, the highest natural enemy counts were in forest transects compared to the interior blueberry orchard (Fig. 2.4). Whereas, in organic orchards, border transects had the highest natural enemy populations (Fig. 2.4). In unmanaged orchards, natural enemies were observed at similar abundance across the three transects (Fig. 2.4).

## Discussion

Our study provides the first characterization of common natural enemy groups found in southeastern subtropical blueberry production. The blueberry agroecosystem in Georgia is one of the fastest growing systems in the region and with an emerging array of pests, this research is ideal and timely for determining the role natural enemies play in pest suppression. Our study demonstrates intensive management practices negatively impact natural enemy abundance and alter natural enemy distribution. Our results also

indicate habitat management may improve natural enemy abundance and potential biocontrol service delivery for southeast systems.

The abundance of natural enemies was significantly different among the three management practices we evaluated. Conventional practices contained the lowest abundance and no accumulation of natural enemies over the season (Fig. 2.3). In addition, some taxa of predatory arthropods were completely absent from conventional fields, yet apparent in both organic and unmanaged blueberry systems (Table 2.2). Studies show significantly higher natural enemy abundance in organic systems (Boutin and Jobin 1998, Koss et al. 2005, Crowder et al. 2010, Martin 2016), and a recent meta-analysis affirms that organic practices increase biodiversity by an average of one-third compared to conventional practices in a variety of crop types with limited information on orchard systems and subtropical regions (Tuck et al. 2014). The current study contributes to lacking information in orchard systems, while also determining the effects within a subtropical region based on extensive field based research. A bioassay study on the lethal effects of registered insecticides in blueberry systems found that reduced-risk insecticides used in organic practices had a lower toxicity to natural enemies than broad-spectrum insecticides used in conventional practices (Roubos et al. 2014). Not only do our findings further support a pattern suggesting intensive management is likely limiting the abundance of natural enemies in managed systems, but we reveal that management practices contain unique distributions of natural enemies.

Intense management practices result in reduced biocontrol services within an agroecosystem due to edge effects, where natural enemies use natural habitat adjacent to agriculture fields for refuge and resources (Rand et al. 2006). In the current blueberry

agroecosystem, the intensive systems support this edge effect of more natural enemies located along the edge of the agroecosystem rather than the interior, similarly depicted in other perennial systems (Picchi et al. 2016, Ingrao et al. 2017). These intensively managed systems can prevent or filter natural enemies from entering the crop interior due to lack of habitat refuge or insecticide application (Schroter and Irmeler 2013). In our conventional systems we found the highest abundance of natural enemies in the forested border, whereas organic systems displayed a tendency for higher natural enemy abundance at the edge of the orchard. Rand et al. (2006) predicted edge effects should be weaker in less intensive systems, which is revealed by the unmanaged systems we found in our blueberry system with an even distribution of natural enemies from the interior to the forested habitat. The ultimate goal for natural pest management is to regulate pests throughout the system, which is influenced by availability of suitable high quality habitat within agricultural landscapes (Landis et al. 2000, Sunderland and Samu 2000, Marshall and Moonen 2002, Blaauw and Isaacs 2015). Our study suggests management intensity alters biocontrol opportunities by redistributing natural enemies within conventional systems, and to a lesser extent in organic systems.

Within field non-crop habitats can alleviate the negative effects of intensive management by providing refuge and resources for natural enemies throughout an agroecosystem (Schellhorn et al. 2014, Amaral et al. 2016, Landis 2016). Previous studies in blueberry systems have examined different ground cover amendments between crop rows revealing bare ground negatively impacts ground dwelling natural enemies (O'neal et al. 2005, Renkema et al. 2012, Jones et al. 2016). While our current study lacks a balanced design of within orchard vegetation compared to orchards without

vegetation in each of the management practices, initial effects on natural enemy patterns were revealed. Vegetation between rows was present in organic and unmanaged systems, whereas conventional systems used herbicides to suppress vegetative growth leading to bare ground between rows. Both herbicide application and mowing affect vegetation structure but to varying degrees; mowing in organic and unmanaged orchards maintains stable vegetation whereas herbicide in conventional orchards removes all vegetation. Infrequent mowing provides additional habitat structure for building predator populations and potential pest suppression (Letourneau et al. 2011). In the blueberry orchards with vegetation between rows, predators were estimated at 8.09 ( $\pm 0.67$ ) per plant compared to the bare ground between blueberry crop rows of 2.14 ( $\pm 0.26$ ). Within field habitat management appears to influence natural enemy presence; however, further study on the combined effects of insecticide applications and habitat management is needed to clarify the interactive roles of these management tactics. These results suggest that enhancing habitat in and around fields through reduced herbicide use and infrequent mowing may improve biocontrol services in southeast blueberry systems. Future experiments will evaluate the effects of varying vegetation between blueberry rows and optimizing habitat and pest management techniques towards attracting diverse natural enemy communities.

Of the major natural enemy groups, generalist predators were the dominant taxa observed in this system and are known to commonly contribute to biological control in agricultural systems (Symondson et al. 2002, Richardson and Hanks 2009, Cardenas et al. 2015, Picchi et al. 2016). Salticidae and Araneidae were the most numerically abundant spider taxa observed across all management practices in this system (Fig. 2.2). Each taxon displays different foraging modes. Salticids, or jumping spiders, are hunting spiders

that utilize a mobile foraging strategy to actively seek out prey with a greater diet breadth than other spider groups (Nyffeler 1999). Araneids, or orb-weaver spiders, are web-weaver spiders that utilize a 'sit-and-wait' strategy and depend on mobile prey that are abundant in the environment (Nyffeler 1999). The two most prevalent insect predators were Reduviidae, which consume prey through piercing-sucking mouthparts, and Carabidae, which consume prey through chewing mouthparts. Within blueberry systems, the combined feeding methods from these four generalist predatory taxa, along with specialist parasitoids, may help regulate a diversity of pests in the environment (Symondson et al. 2002). Foraging tactics vary among these groups, as each mode of feeding potentially partitions ecological niches in the environment (Gable et al. 2012). Continuing research will aim at uncovering feeding links between predators and key pests to determine which taxa effectively contribute to biological control. Information on predator diet will provide a foundation of knowledge to understand and promote biological control options in blueberry production.

Overall, our study expands knowledge on natural enemies associated with southeastern subtropical U.S. blueberry agroecosystems and encourages the use of ecologically based management systems. Building ecologically based management systems requires comprehensive knowledge of the natural enemies present and the biological control services they provide. With the information provided in our study, we have identified the dominant predator taxa and can discern the impacts of management practice on natural enemy abundance and distribution. Our findings suggest the potential for production systems that mimic unmanaged systems to promote recolonization by natural enemies and possibly improve biological control.

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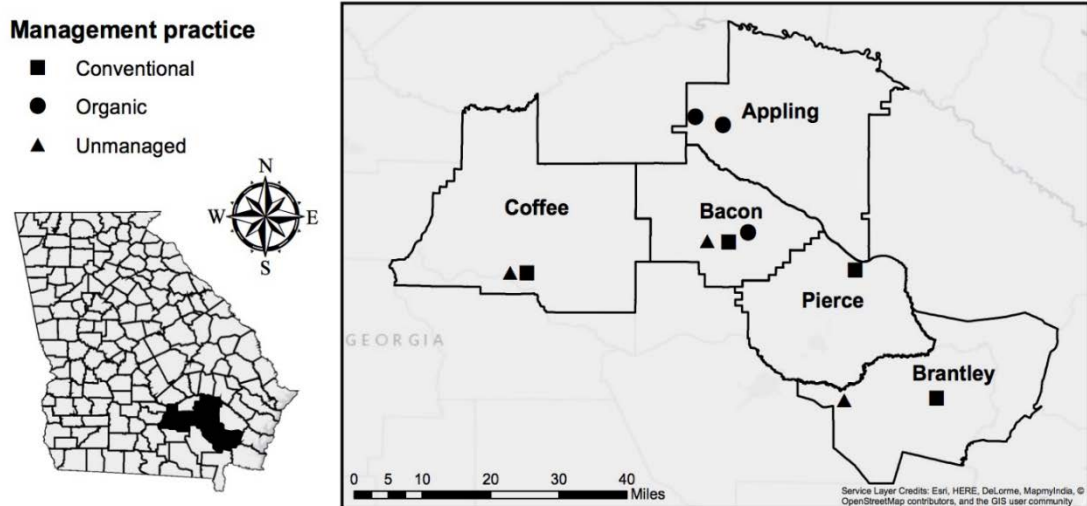
**Table 2.1.** Descriptive summary of taxa collected from suction sampling throughout the 2016 study. Percentages represent the proportion of each functional group per management practice and values represent the mean ( $\pm$  1SE) of each taxon observed over the season for all sites in the respective management practice. Total represents the counts pooled over spatial-temporal extent of study for all taxa observed.

Taxa	Conventional	Organic	Unmanaged	Total
<b>Arachnids</b>	78%	73%	78%	
Araneae				
Salticidae	1.31 $\pm$ 0.27	3.60 $\pm$ 1.08	5.46 $\pm$ 0.96	219
Immature spiders	1.19 $\pm$ 0.24	2.00 $\pm$ 0.60	5.04 $\pm$ 0.80	182
Araneidae	0.73 $\pm$ 0.15	2.47 $\pm$ 0.46	3.92 $\pm$ 0.67	150
Oxyopidae	0.46 $\pm$ 0.15	0.47 $\pm$ 0.22	3.04 $\pm$ 0.58	92
Thomisidae	0.42 $\pm$ 0.11	1.40 $\pm$ 0.38	1.88 $\pm$ 0.30	77
Lycosidae	0.19 $\pm$ 0.10	0.20 $\pm$ 0.11	1.25 $\pm$ 0.33	38
Linyphiidae	0.15 $\pm$ 0.07	0.27 $\pm$ 0.15	0.92 $\pm$ 0.25	30
Clubionidae	0.12 $\pm$ 0.06	0.53 $\pm$ 0.22	0.67 $\pm$ 0.21	27
Tetragnathidae	0.27 $\pm$ 0.09	0.13 $\pm$ 0.09	0.50 $\pm$ 0.18	21
Theridiidae	0.12 $\pm$ 0.06	0.13 $\pm$ 0.09	0.29 $\pm$ 0.11	12
Ulobridae	0.00 $\pm$ 0.00	0.00 $\pm$ 0.00	0.08 $\pm$ 0.06	2
Gnaphosidae	0.04 $\pm$ 0.04	0.07 $\pm$ 0.07	0.00 $\pm$ 0.00	2
Opiliones				
Phalangidae	0.00 $\pm$ 0.00	0.07 $\pm$ 0.17	0.06 $\pm$ 0.08	5
<b>Insect predators</b>	12%	12%	11%	
Reduviidae	0.38 $\pm$ 0.14	0.67 $\pm$ 0.47	0.83 $\pm$ 0.26	40
Carabidae	0.08 $\pm$ 0.05	0.33 $\pm$ 0.21	0.92 $\pm$ 0.32	29
Vespidae	0.12 $\pm$ 0.06	0.27 $\pm$ 0.12	0.79 $\pm$ 0.29	26
Coccinellidae	0.08 $\pm$ 0.08	0.00 $\pm$ 0.00	0.46 $\pm$ 0.19	13
Syrphidae	0.00 $\pm$ 0.00	0.33 $\pm$ 0.16	0.13 $\pm$ 0.07	8
Chrysopidae	0.12 $\pm$ 0.06	0.20 $\pm$ 0.14	0.04 $\pm$ 0.04	7
Mantidae	0.00 $\pm$ 0.00	0.00 $\pm$ 0.00	0.08 $\pm$ 0.06	2
Anthocoridae	0.00 $\pm$ 0.00	0.07 $\pm$ 0.07	0.04 $\pm$ 0.04	2
Geocoridae	0.00 $\pm$ 0.00	0.00 $\pm$ 0.00	0.04 $\pm$ 0.04	1
<b>Parasitoids</b>	10%	15%	11%	
Parasitica	0.65 $\pm$ 0.22	2.27 $\pm$ 0.64	3.21 $\pm$ 0.93	128

**Table 2.2.** Analysis of natural enemy population variability in blueberry agroecosystems.

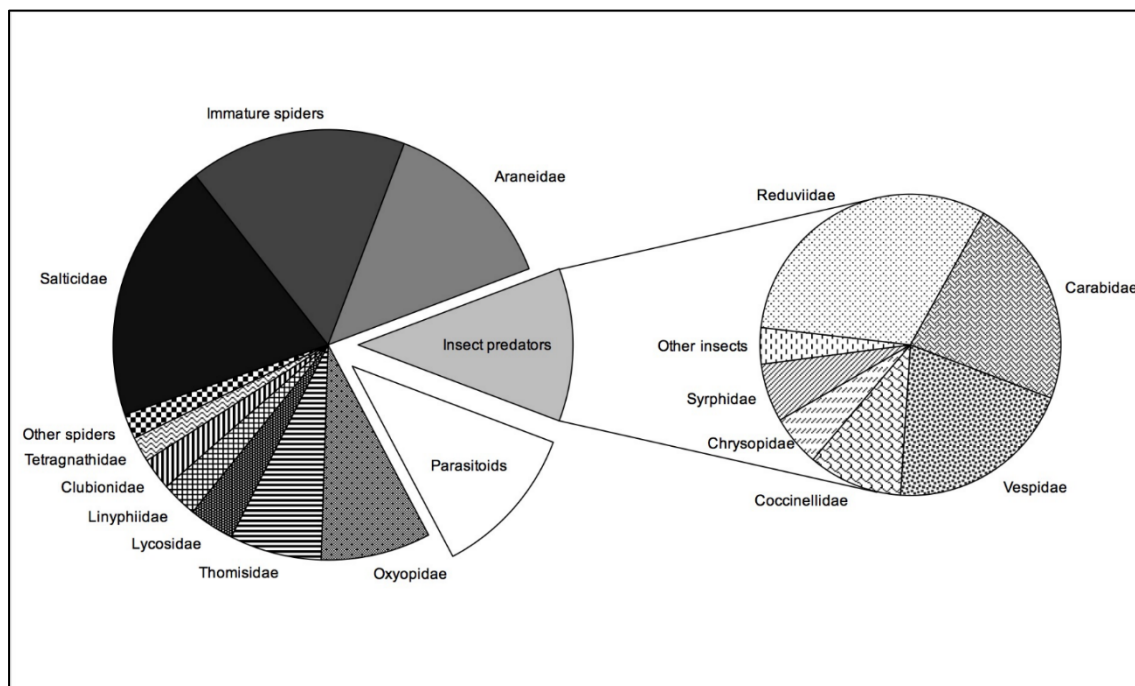
Response variable used was total natural enemy abundance combining all natural enemy taxa. Analysis model was a Generalized Least Squares (gls) fitted with REML to account for repeated measures of communities within orchard overtime (i.e. random error partitioned with date nested within orchard). Table summarizes fixed effects included in the analysis. Sampling date was entered as a continuous variable, and natural enemy abundance was natural logarithm transformed prior to analysis. Variables connected by “:” represent interaction terms.

Variable	df	F-value	P-value
Sampling date	1	0.25	0.6203
Management practice	3	19.46	<b>&lt;0.0001</b>
Transect	2	3.48	<b>0.0330</b>
Orchard size	1	0.04	0.8417
Sampling date:Management practice	2	3.64	<b>0.0281</b>
Management practice:Transect	4	1.03	0.3909
Management practice:Size	2	0.03	0.9713
Residual	180		

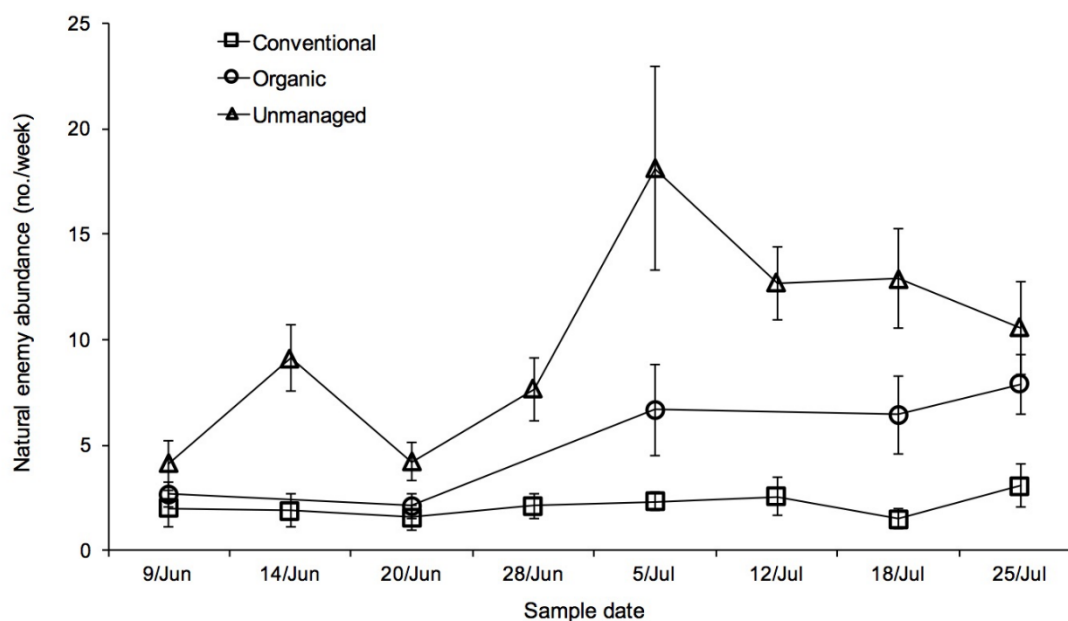


**Fig. 2.1.** Map of research sites sampled in Georgia, USA with symbols to indicate distribution of management practices.

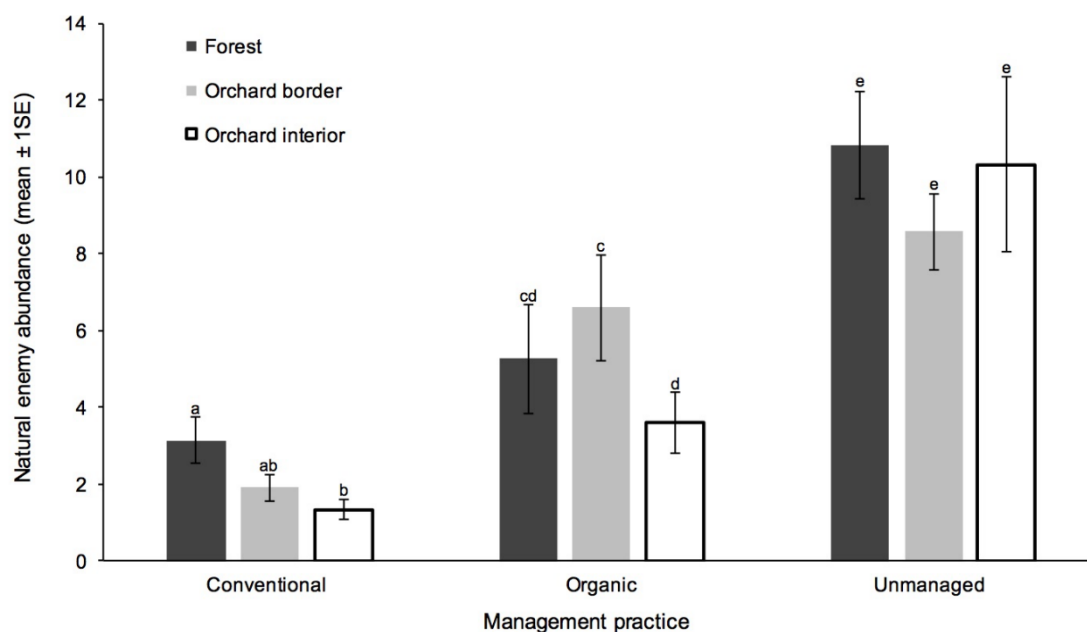




**Fig. 2.2.** Natural enemy composition in blueberry orchards estimated from suction samples pooled over all samples. Primary pie chart represents the proportion of each taxa calculated from total counts ( $N = 1113$ ) with insect predators separated into second pie chart.



**Fig. 2.3.** Seasonal mean ( $\pm 1$ SE) natural enemy abundance from suction samples pooled across transect and site to over time. Symbols represent corresponding management practice where samples were collected during the blueberry growing and harvest season.



**Fig. 2.4.** Mean ( $\pm$  1SE) natural enemy abundance from suction samples pooled across weekly sampling period and site. Bars represent the sampling location (forested habitat, canopy of the blueberry orchard border, and canopy of the blueberry orchard interior) in the respective management practice (conventional, organic, and unmanaged).

CHAPTER 3

RELATIVE IMPORTANCE OF LOCAL AND LANDSCAPE FACTORS IN  
BLUEBERRY AGROECOSYSTEMS: DIFFERENTIAL EFFECTS BETWEEN  
NATURAL ENEMIES AND THE PRIMARY PEST

## Abstract

Natural enemies provide biological control services in agroecosystems, yet the manipulation of natural enemies through both habitat management and the surrounding landscape are often left unexplored. In order to enhance biocontrol services, research must explore how local and landscape factors are affecting the distribution of natural enemies. Our study system included 20 blueberry orchards during the 2017 harvest season (May to July) across the major blueberry producing counties in Georgia, U.S.A. Arthropod collection was performed through suction sampling for natural enemies and sugar-yeast based traps for *Drosophila suzukii*. The results revealed varying effects of local factors at each orchard (e.g., management practice and vegetation presence) and landscape factors surrounding each orchard (e.g., landscape diversity, landscape configuration, and habitat amount). While natural enemies were negatively impacted by conventional management practices at the local scale, the presence of vegetation alleviated these effects by promoting the recolonization of natural enemies. At the landscape scale, habitat diversity played the largest role in increasing natural enemies and habitat configuration had the largest impact on the increase in *D. suzukii*. This research highlights differential effects involving local and landscape factors, while suggesting the most beneficial management strategies. The implementation of habitat management to establish vegetation between crop rows and landscape management towards habitat diversity have the potential to enhance the recolonization of natural enemies within the agricultural system and improve agricultural sustainability.

**Key Words** Biological control, *Drosophila suzukii*, ecosystem services, landscape, local vegetation, spider, spotted wing drosophila.

## Introduction

In recent years, insect biomass has declined significantly causing great concern for the ecosystem services they provide (Hallmann et al. 2017). Particularly, biological control services are estimated at \$13 billion per year in the U.S. (Losey and Vaughan 2006), in which many agricultural systems rely on for pest suppression. Yet, agricultural intensification from chemical control and simplification of landscapes are currently diminishing those ecosystem services (Tscharntke et al. 2005, Thies et al. 2011, Bommarco et al. 2013, Rusch et al. 2016). In order to alleviate the negative effects of agricultural intensification and promote ecosystem services, we must identify the various environmental factors that drive arthropod abundances found in agroecosystems (Liere et al. 2017).

Those environmental factors include local and landscape effects. The local factors are defined by immediate effects that occur within the agricultural system, such as management practice or insecticide regime, and maintenance of vegetation (Chaplin-Kramer and Kremen 2012). Landowners can more easily manipulate these local factors by utilizing habitat management techniques. There is empirical evidence that habitat management through the implementation of vegetation within agricultural systems benefits natural enemies by providing resources and refuge (Landis et al. 2000, Boller et al. 2004, Fiedler et al. 2008, Thomson and Hoffmann 2010, Paredes et al. 2015). However, these local effects are nested within the surrounding landscape and the success of habitat management may depend on the landscape factors present. Landscape factors are defined by gradual effects that take place at a larger scale, which incorporate the composition and configuration of the landscape (Fahrig et al. 2011). The composition of

the landscape includes the proportions of different land cover types and the availability of non-crop habitat within the surrounding ecosystem. The configuration of the landscape includes the spatial arrangement and complexity of cover types (Veres et al. 2013). A certain composition or type of habitat may support some arthropods more than others, whereas a more complex configuration of the habitat will support a greater diversity of arthropods (Schmidt and Tscharntke 2005 and Sarthou et al. 2014). Landscape management through designing the surrounding agroecosystem structure towards higher diversity of suitable habitat has also shown to positively impact natural enemy abundances (Langellotto et al. 2004, Veres et al. 2013 and Rusch et al. 2012). Therefore, it is understood that both local and landscape factors may contribute to the recruitment of natural enemies, however the impact on primary pests in agroecosystems are often overlooked (Weibull et al. 2003, Chaplin-Kramer and Kremen 2012).

In recent reviews of the interactions between arthropod abundance and environmental factors, 36 of the 38 selected studies investigated annual cropping systems (Bianchi et al. 2006 and Rusch et al. 2016). Fewer studies have investigated perennial systems, which are believed to have different arthropod interactions compared to annual systems due to the year round stability of vegetation within the agricultural system. For instance, natural enemies in the vineyards and cherry trees did not respond to higher availability of non-crop habitat (Stutz and Entling 2011 and Alberto et al. 2012), yet higher landscape diversity in olive orchards caused a reduction in the primary pest of concern (Ortega and Pascual 2014). The information provided from the current blueberry system will contribute to a knowledge gap of environmental factors in more stable, perennial systems. Here, we assess the relative importance of local and landscape factors

on both natural enemies and the primary pest. The primary, invasive pest we investigate is *Drosophila suzukii*, or spotted wing drosophila, which infests a range of cultivated and wild fruit hosts (Hauser 2011 and Lee et al. 2015). The wide host use of *D. suzukii* has allowed this pest to successfully migrate between landscapes as there is an abundance and diversity of fruit host resources available in the southeastern U.S. region (Grant and Sial 2016). Multiple generations of *D. suzukii* can then disperse between the adjacent forest to within blueberry orchards (Haviland et al. 2016, Klick et al. 2016b), which creates a unique challenge for pest management strategies. Therefore, the current blueberry system in the southeastern U.S. and its recent invasion of *D. suzukii* provide an interesting model system to evaluate the local and landscape factors that contribute to natural enemy and pest abundances. The current study aims to determine which factors may increase natural enemies and to predict landscape conditions that impact the invasive blueberry pest, *D. suzukii*.

Our research addresses three key questions: (1) how do local and landscape factors affect the distribution of natural enemies and the primary pest in blueberry agroecosystems? (2) Which environmental factors have the greatest relative importance for shaping arthropod abundances? (3) And how may this information be incorporated into habitat or landscape management practices to promote the delivery of biological control services? We expect that increased local habitat resources and increased landscape diversity will have beneficial results on natural enemy abundances, which may directly limit pest abundances as well. Our approach compares a gradient of different local and landscape effects in order to better understand how arthropods are shaped in the agroecosystem. These findings will help to suggest methods of optimizing biological



control services and the response of arthropods within a perennial system will provide a unique perspective to help explain how we may improve agricultural sustainability across landscapes.

## Materials and Methods

**Study Area.** In 2017, using the University of Georgia extension network, 20 commercial blueberry orchards were selected in seven counties across South Georgia, U.S.A. (Coffee Co., Bacon Co., Appling Co., Pierce Co., Brantley Co., Ware Co., and Jeff Davis Co.; Fig. 3.1). Blueberry production in the region harvests *Vaccinium ashei* (rabbiteye blueberry) and *Vaccinium corymbosum* (southern highbush blueberry). Our study area included a spatial extent covering approximately 8,846 km<sup>2</sup>. Blueberry orchards were selected to vary in both local management and surrounding landscape. We attempted to select a balanced number of management practices separated by at least 1 km, except two sites separated by 450 m. The different management systems among blueberry orchards included conventional (12), certified organic (5), and unmanaged (3) (Fig. 3.1). Management classifications were made based on intensity and type of insecticides applied. The twelve conventional sites utilized broad-spectrum synthetic insecticides, including primarily organophosphates and pyrethroids, and in some cases a spinosyn (i.e., Delegate<sup>TM</sup>), and herbicides often applied between the blueberry rows for weed management. The five organic sites utilized reduced-risk organically certified (OMRI listed) insecticides and herbicides were not applied between blueberry rows. The three unmanaged sites varied from abandoned orchards to small scale harvesting with vegetation present between orchard rows mowed infrequently. The surrounding

landscape at each site varied along a gradient in habitat composition and configuration. The 2016 USDA cropland data layer provided land use information, which could then be grouped into six categories comprised of annual crops, perennial crops, forest, seminatural habitat, urban areas, and water bodies (Fig. 3.1).

**Sampling.** Each orchard site was sampled every other week along a transect containing three stations, similar to methods described in Whitehouse et al. (2017), for a total of 60 samples per period. Transects included locations of 25 m into the crop (orchard interior), along the orchard margin or first crop row (orchard border), and 15 m within the adjacent, non-crop forested habitat (forest) (Fig. 3.1). At each location, a 60-second suction sample method was used to collect natural enemies with a modified reversed-flow leaf blower (SH 86 C-E; Stihl, Waiblingen, Germany). To prevent sampling bias caused by different vegetation structure (Hossain et al. 1999), the sampling was conducted within a 2 m<sup>2</sup> area in the blueberry orchard and forested region. The samples were transferred to plastic bags and placed on ice until permanent storage at -20 °C in the laboratory. Individual specimens were separated from the collected samples, identified to taxonomical level (family or order), and preserved in 95% ethanol at -20 °C towards future molecular analysis. Along similar transects at each site, spotted wing drosophila yeast-sugar bait traps were deployed to monitor pest pressure, which is the accepted method of trapping *D. suzukii* (Landolt et al. 2012). After the two-week period, bait traps were collected and transferred to the University of Georgia Blueberry Research and Demonstration Farm located in Alma, GA. Male and female *D. suzukii* were then separated from the bait traps and recorded.

**Local and landscape factors.** Observations at each orchard were used to calculate the various local factors. The characteristics measured at each orchard include the management practice (conventional, organic, unmanaged), sample period (date during sample collection), and vegetation height (measured between blueberry rows during sample collection; m). The landscape was characterized using Geographic Information Systems (ArcGIS, version 10.3.1; Environmental Systems Research Institute, Redlands, California, USA) to create polygon maps based on Landsat world imagery (ESRI 2016) and regional land use maps (United States Department of Agriculture; Cropland Data Layer 2016) (Fig. 3.1). A radius of 1 km supports the strongest interaction between predators (i.e. spiders) and landscape (Schmidt et al. 2008), therefore we created consistent buffers at each site among all orchards (Fig. 3.1). The landscape characteristics measured at each orchard include the habitat amount (percentage of non-crop habitat), landscape diversity (Shannon habitat diversity index; SHDI), and landscape configuration (edge density; ED). SHDI and ED values were calculated using a spatial pattern analysis program for categorical and continuous maps (FRAGSTATS, version 4; University of Massachusetts, Amherst).

**Analysis.** The analyses were performed using R version 3.3.1 (R Core Team 2016), and significant main effects were analyzed with linear contrasts for multiple comparisons of mean responses. Data over the May-July sampling period included a total of 420 observations to determine the effect on the response variables, natural enemies and *D. suzukii*. Fixed effects included the local (management practice, sample period, and vegetation height) and landscape factors (habitat amount, landscape diversity, and

landscape configuration). For each response variable, 7 models for local factors and 7 models for landscape factors were fitted for both response variables. Linear models (using “lm”: function in R version 3.3.1) were used to assess the effects on natural enemies and *D. suzukii* per respective fixed effect model. We built all possible combinations of fixed factors (management practice, vegetation height, sampling period, Shannon Habitat Diversity Index [SHDI], Edge Density [ED], and proportion of non-crop habitat) and response variables (Natural enemies and *D. suzukii*). A total of 28 models were compared using the Akaike Information Criterion (AIC). Models with a difference in  $AIC > 2$  suggested these were non-competing models that could be omitted from the analysis.

## Results

We collected a total of 2,292 natural enemies from a total of 420 suction samples including: 1,896 arachnids, 197 insect predators, and 199 parasitoids. Arachnids made up of Araneae, the true spiders, combined with Opiliones, harvestmen, were the numerically dominant predatory arthropod taxa. Insect predators comprised ten families, and Hymenopteran parasitoids comprised parasitic Apocrita. The distribution of Arachnids as the dominate predator taxa was consistent across all three management practices. A total of 3,036 spotted wing drosophila (*Drosophila suzukii*) individuals were collected from 420 deployed sugar-yeast bait traps.

Model selection for natural enemy abundance indicated there were three competing models when comparing local factors (mp + per; mp; mp + per + veg) and only one competing model when comparing landscape factors (ed + prop). The lowest

AIC value identified management practice and sampling period to be the best fitting model for local effects on natural enemy abundance (Table 3.1). The lowest AIC value identified edge density and proportion of non-crop habitat to be the best fitting model for natural enemy abundance at the landscape scale (Table 3.1). Sample period, management practice, vegetation, non-crop, and SHDI contribute to explaining variability of natural enemy populations in blueberry orchards (Table 3.2). Based on the best fitting AIC models, the local interaction of sample period and management practice resulted in a significant difference (p-value=0.007; Table 3.2). The landscape interaction of non-crop and edge density also resulted in a significant difference (p-value<0.0001; Table 3.2).

Model selection for *D. suzukii* abundance indicated there were two competing models when comparing local factors (mp + per; mp + per + veg) and three competing model when comparing landscape factors (ed + prop; shdi + prop; ed + shdi +prop). The lowest AIC value identified management practice and sampling period to be the best fitting model for local effects on *D. suzukii* abundance (Table 3.3). The lowest AIC value identified edge density and proportion of non-crop habitat to be the best fitting model for *D. suzukii* abundance at the landscape scale (Table 3.3). Sample period, management practice, non-crop, and edge density contribute to explaining variability of *D. suzukii* populations in blueberry orchards (Table 3.4). Based on the best fitting AIC models, the local interaction of sample period and management practice resulted in a significant difference (p-value<0.0001; Table 3.4). The landscape interaction of non-crop and edge density did not result in a significant difference.

The following results demonstrate the effect of local factors on arthropod abundance at each blueberry orchard site. Arthropod accumulation over time in orchards

was dependent upon management practice (Fig. 3.2). There was a significant interaction between management practice and sampling period on natural enemies ( $F_{12, 419} = 2.31$ ,  $p = 0.007$ ; Table 3.3) and no significant interaction on *D. suzukii* ( $F_{12, 419} = 1.002$ ,  $p = 0.446$ ; Table 3.4). Orchard transects within management practices also showed significant differences in the natural enemy populations (Fig. 3.3). In conventional orchards, the highest natural enemy counts were in forest transects ( $6.699 \pm 0.521$ ) compared to the orchard border ( $2.831 \pm 0.323$ ) and the orchard interior ( $1.940 \pm 0.220$ ). In organic orchards, the forest contained the highest natural enemy count as well ( $10.314 \pm 1.246$ ) compared to the orchard border ( $7.143 \pm 0.943$ ) and orchard interior ( $5.343 \pm 0.864$ ). In unmanaged orchards, natural enemies were observed at similar abundance across from within the adjacent forest ( $8.364 \pm 1.325$ ), orchard border ( $7.864 \pm 1.501$ ), and orchard interior ( $8.409 \pm 1.09$ ). Similar interactions were found in *D. suzukii* populations from management practice and within orchard transects (Fig. 3.3). In conventional orchards, the highest *D. suzukii* counts were in the forest transects ( $16.638 \pm 4.264$ ) compared to the orchard border ( $3.694 \pm 1.084$ ) and orchard interior ( $3.12 \pm 0.633$ ). In organic orchards, the forest ( $9.031 \pm 3.448$ ) and orchard border ( $8.206 \pm 4.457$ ) were both higher than the orchard interior  $3.091 \pm 1.254$ ). In unmanaged systems, the orchard interior ( $14.381 \pm 3.526$ ) contained the highest *D. suzukii* abundance compared to the orchard border ( $6.048 \pm 1.487$ ) and within the forest ( $5.579 \pm 2.173$ ). During the local vegetation analysis (Fig. 3.4) we found that conventional systems with vegetation between blueberry rows had a significantly higher abundance of natural enemies ( $2.688 \pm 0.398$ ) compared to bare ground between blueberry rows ( $1.519 \pm 0.239$ ). There was no

significant difference between the two treatments in *D. suzukii* abundance from vegetation ( $3.355 \pm 0.995$ ) and bare ( $2.955 \pm 0.830$ ).

The following results demonstrate the effect of landscape factors on arthropod abundance at each blueberry orchard site. Natural enemies significantly decreased with a higher percentage of non-crop habitat ( $F_{1, 419} = 29.03$ ,  $p < 0.0001$ ) and significantly increased with a higher SHDI, habitat diversity ( $F_{12, 419} = 1.002$ ,  $p = 0.446$ ), while there was no significant difference based on ED, habitat complexity (Fig. 3.5). The abundance of *D. suzukii* significantly increased with higher percentage of non-crop habitat ( $F_{1, 419} = 14.44$ ,  $p < 0.005$ ) and higher ED, habitat complexity, ( $F_{1, 419} = 14.89$ ,  $p < 0.005$ ), while no significant difference based on SHDI habitat diversity (Fig. 3.5).

## Discussion

Our study provides the first analysis of the local and landscape effects on arthropods found within a blueberry agroecosystem. It has been proven that the current blueberry system is not utilizing the full potential of biocontrol services as there is a significant reduction in the abundance of natural enemies found in conventional orchards (Whitehouse et al. 2017). The current study expands on this finding with a more holistic approach to determine the effects of various environmental factors that play a role in shaping not only natural enemies, but also the primary pest of concern. The results indicate the relative importance of local and landscape factors, while suggesting management techniques to promote natural enemies and potentially improve pest suppression.

At the local scale, natural enemies are reduced in conventional systems, which merits the need to evaluate other local variables that may improve sustainability within orchards (Desneux et al. 2007 and Roubos et al. 2014). The presence of local vegetation may do just that, where orchards that maintain vegetation between blueberry rows increased natural enemy abundances, whereas *D. suzukii* remained unaffected (Fig. 3.3). This local effect of vegetation within the cropping system is widely believed to increase both abundance and diversity of natural enemies (Aviron et al. 2016, Phillips and Gardiner 2016, Lemessa et al. 2015, Koh et al. 2015, Chaplin-Kramer et al. 2012, Thomson and Hoffmann. 2010). The vegetation provides resources and refuge to promote the recolonization of natural enemies (Schellhorn et al. 2014), and previous studies in blueberry systems have examined the benefit of ground cover amendments between crop rows (O'neal et al. 2005, Renkema et al. 2012, Jones et al. 2016). While our study further contributes to the benefit for natural enemies, an important finding is the neutral effect on the primary pest. *D. suzukii* was not significantly different between the bare ground or vegetation treatments in conventional systems, therefore local vegetation may be incorporated into current management practices without repercussions of *D. suzukii* increase. Habitat management through the availability of suitable high quality habitat within agricultural landscapes may then improve the delivery of ecosystem services (Landis et al. 2000, Sunderland and Samu 2000, Marshall and Moonen 2002, Blaauw and Isaacs 2015) and may become a realistic tool to help suppress large pest outbreaks. The adoption of local vegetation by growers will not only reduce cost of herbicide applications, but also increase potential biological control services provided by natural enemies.



Predator-prey interactions vary depending on the mobility of taxa within the environment (Fagan et al. 1999, Rand et al. 2006, Macfadyen and Muller 2013), therefore we assess the distribution of arthropods within the agroecosystem. Conventional systems showed a concentration of natural enemies towards the adjacent forest, similar to *D. suzukii* distribution. However, there are stark differences between natural enemies and *D. suzukii* in unmanaged orchards. The distribution of *D. suzukii* was significantly higher in the forested regions of conventional orchards and significantly higher in the orchard interior of the unmanaged orchards (Fig. 3.3). This is most likely due to the ability of *D. suzukii* to utilize a large range of non-crop hosts found natively in the southeastern U.S. (Lee et al. 2015, Arno et al. 2016). The resiliency of *D. suzukii* creates a unique challenge for pest management where this pest can successfully migrate between different habitats over multiple generations from the adjacent forest to blueberry orchards (Haviland et al. 2016, Klick et al. 2016b). Survey and mark-recapture methods have documented the dispersal of *D. suzukii* between habitats (Klick et al. 2016a), and the current study also highlights the movement of this pest from the forest to blueberry crop comparing conventional and unmanaged orchards. Field observations at the unmanaged orchards located within the current study affirm the presence of both abandoned blueberry (*Vaccinium* spp.) and native, uncultivated fruit including blackberries (*Rubus* spp.) and pokeweed (*Prunus avium*), which are highly susceptible hosts for *D. suzukii* (Lee et al. 2015 and Klick et al. 2016b). Without exposure to local disturbances, such as insecticides, in the unmanaged orchards, *D. suzukii* is capable of migrating with ease between habitats. And with the larger abundance situated within the orchard interior than the adjacent forest in unmanaged orchards, we find that *D. suzukii* prefer such conditions

with ample hosts. As a consequence, the high mobility of this pest urges the need to compare how arthropods are utilizing the resources in the surrounding landscape.

At the landscape scale, an increase in non-crop habitat caused a decrease in natural enemy abundance and an increase in *D. suzukii* abundance (Fig. 3.5A). While this calculation may be useful, the natural landscape is not simply divided into two categories and the non-crop habitat is made up of various habitat types (Shackelford et al. 2013 and Jankovic et al. 2017). For this reason, we stress the importance of assessing the habitat diversity and complexity within the landscape. We find that natural enemy abundance significantly improved with an increase in landscape diversity (SHDI), while *D. suzukii* had no significant effect (Fig. 3.5B). Alternatively, *D. suzukii* abundance significantly increased with an increase in habitat complexity (ED) and natural enemies had no significant effect (Fig. 3.5C). Here, we find differential effects on two groups of arthropods that may utilize different resources in the surrounding landscape. For instance, generalist species may benefit from enhanced landscape diversity, unlike more specialist species that cannot utilize the variety of resources in a diverse landscape (Krauss et al. 2003 and Schmidt et al. 2008). The current study evaluates the response of natural enemies as a whole, which may contain either positive or negative on particular species within the group. Despite this fact, we sought to determine the effect on natural enemies as a whole and found that they respond favorably to increased diversity in the landscape, unlike *D. suzukii* that benefits from increased complexity surrounding the landscape. So the success of local vegetation to promote natural enemies depends upon the habitat diversity surrounding each orchard. Furthermore, the *D. suzukii* abundance requiring higher habitat complexity corresponds with the results regarding *D. suzukii* distribution in

the agroecosystem, as this pest successfully migrates between the forested regions and blueberry orchards. Our aim was to assess the differential effects between two major groups of taxa, natural enemies and the primary pest, thus opening the discussion to potential landscape management tactics.

Many studies analyze natural enemies without considering the alterations of the key pests in the system (Gardiner et al. 2009, Jonsson et al. 2015, Lemessa et al. 2015, Paredes et al. 2015, Zhao et al. 2016), which is necessary to calculate the tradeoffs of incorporating new management tools (Bianchi et al. 2006, Veres et al. 2013, Rusch et al. 2016). A review by Veres et al. (2013) attempted to uncover the effects of landscape factors on pest control by natural enemies and urges the need for specific interactions of natural enemies and associated pests in their respective cropping system. Although the consensus of literature illustrates the increase of non-crop habitat and higher landscape complexity causes an increase in natural enemy abundance (Tscharntke et al. 2005, Chaplin-Kramer et al. 2011, Veres et al. 2013, Aviron et al. 2016, Landis 2016), there is little information regarding the environmental factors contributing to pest abundances. Simplified landscapes prompt the weakening of predator-prey interactions (Kruess and Tscharntke 1994, Rusch et al. 2016), therefore landowners should consider the benefits of manipulating the surrounding landscape. For example, landscape management is a long-term approach to improve sustainable crop production, while maximizing the biocontrol services delivered in the landscape (Woltz et al. 2012 and Veres et al. 2013). There is growing support to demonstrate that landscape diversity can cause pests in cropping systems to decrease due to increased pressure of biocontrol agents (Bianchi et al. 2006, Gardiner et al. 2009, Vasseur et al. 2013 and Rusch et al. 2016). For instance, Picchi et al.

(2016) reported a decrease in the olive fruit fly pest (*Bactrocera oleae*), which was likely caused by an increased spider abundance found in more diverse landscapes. However, the primary pest investigated in the current study is able to use a wide range of non-crop hosts in more abundant habitats, therefore we are uncertain that natural enemies may provide effective biological control services to suppress the invasion of *D. suzukii*.

Although the current study evaluates natural enemies and *D. suzukii*, other known blueberry pests may also be affected by the environmental factors examined. Due to our sampling period taking place during the harvest season, there were low secondary pest abundances from vacuum sampling and yellow sticky card traps collected at the blueberry orchard sites. And prior to conducting our research, the examined blueberry region experienced a frost event during the blueberry flower bud stages, ultimately causing a decrease in yields, altered management practices, and potential disruption of arthropod communities. As a result, we recommend that future research utilize a long-term approach to determine the relative importance of environmental factors throughout the year, while considering the effects on all key pests present in the agroecosystem. Further research on direct feeding links between predator taxa and key pests through molecular gut-content analysis will also provide vital information for management tactics to enhance predator taxa that effectively contribute to biological pest control.

Overall, our results indicate that local and landscape factors have differential effects on natural enemies and the primary pest in the study system. Local factors shape arthropod abundances where natural enemies and *D. suzukii* decreased from conventional practices, however the presence of vegetation in conventional systems alleviated those negative effects by promoting natural enemies within the agricultural system. We suggest

the incorporation of habitat management; however, the recolonization of natural enemies depends on the landscape factors present in the system. At the landscape scale, natural enemies respond favorably to increased diversity in the landscape, yet *D. suzukii* benefitted from increased configuration surrounding the landscape. *D. suzukii* abundance and distribution highlighted in this study infers the success of this pest to utilize resources across the landscape. Thus, considerations to evaluate the response of the primary pest in the agroecosystem should be made when designing landscapes. The information provided in our study furthers our understanding of the differential effects of environmental factors that drive natural enemies and the primary pest investigated. Given the importance of both habitat and landscape management, the adoption of ecologically-based management systems across multiple agroecosystems has the potential to greatly enhance ecosystem services provided by natural enemies.

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**Table 3.1.** Results of model selection comparing combinations of predictor variables explaining variation in natural enemy abundance in blueberry orchards. Models were ranked in relation to fitted (lm) model reported with all relevant interactions for the experimental design. The best fitted model is represented by the lowest AIC and other models are ranked in relation to change in AIC. Predictor variables are abbreviated as: management practice (mp), sample period (per), vegetation (veg), edge density (ed), shannon habitat diversity index (shdi), and proportion of non-crop (prop).

Local effects model	$\Delta$ AIC
mp + per	0
mp	0.653
mp + per + veg	1.851
mp + veg	2.155
veg + per	31.72
veg	34.41
per	61.53
Landscape effects model	$\Delta$ AIC
ed + prop	0
shdi + prop	16.93
shdi	17.159
ed + shdi	17.406
ed + shdi + prop	18.68
prop	31.08
ed	43.58

**Table 3.2.** Analysis of natural enemy population in blueberry agroecosystems. Response variable was total natural enemy abundance combining all natural enemy taxa. Analysis of Variance (ANOVA) with a significance level of  $P < 0.05$ . Significant main effects were analyzed with Tukey HSD for multiple comparisons of mean responses.

Variable	df	F-value	P-value
<b>Local effects</b>			
Sample period	6	3.293	<b>&lt;0.005</b>
Management practice	2	34.98	<b>&lt;0.0001</b>
Vegetation	1	30.29	<b>&lt;0.0001</b>
Sample period:Management practice	12	2.31	<b>0.007</b>
<b>Landscape effects</b>			
Non-crop	1	14.46	<b>&lt;0.0001</b>
Edge density	1	1.779	0.183
SHDI	1	29.03	<b>&lt;0.0001</b>
Non-crop:Edge density	1	32.721	<b>&lt;0.0001</b>

**Table 3.3.** Results of model selection comparing combinations of predictor variables explaining variation in *Drosophila suzukii* abundance in blueberry orchards. Models were ranked in relation to fitted (lm) model reported with all relevant interactions for the experimental design. The best fitted model is represented by the lowest AIC and other models are ranked in relation to change in AIC. Predictor variables are abbreviated as: management practice (mp), sample period (per), vegetation (veg), edge density (ed), shannon habitat diversity index (shdi), and proportion of non-crop (prop).

Local effects model	$\Delta$ AIC
mp + per	0
mp + per + veg	0.711
veg + per	6.479
per	9.327
mp + veg	11.161
mp	15.623
veg	20.435
Landscape effects model	$\Delta$ AIC
ed + prop	0
shdi + prop	0.23
ed + shdi + prop	0.564
ed	6.19
ed + shdi	6.826
prop	6.911
shdi	20.278



**Table 3.4.** Analysis of *Drosophila suzukii* population in blueberry agroecosystems.

Response variable was total *Drosophila suzukii* abundance combining all natural enemy taxa. Analysis of Variance (ANOVA) with a significance level of  $P < 0.05$ . Significant main effects were analyzed with Tukey HSD for multiple comparisons of mean responses

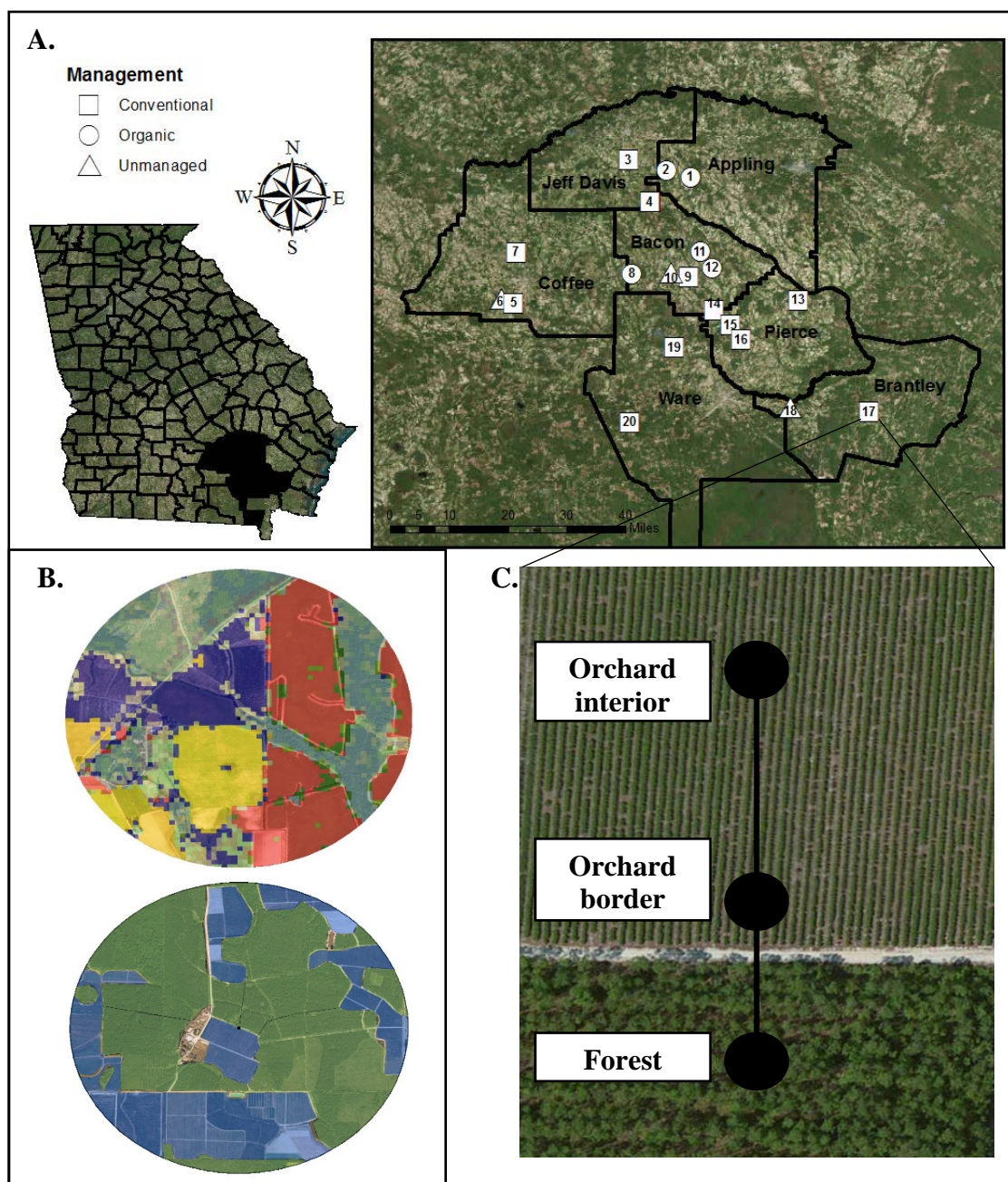
Variable	df	F-value	P-value
<b>Local effects</b>			
Sample period	6	7.38	<b>&lt;0.0001</b>
Management practice	2	6.492	<b>&lt;0.005</b>
Vegetation	1	3.064	0.081
Sample period:Management practice	12	1.002	0.446
<b>Landscape effects</b>			
Non-crop	1	14.44	<b>&lt;0.005</b>
Edge density	1	14.89	<b>&lt;0.005</b>
SHDI	1	0.607	0.436
Non-crop:Edge density	1	2.536	0.112

**Table 3.5.** Local factors at all 20 blueberry orchard sites with corresponding arthropod mean abundance during each sampling period. Management practice was based on chemical control inputs and vegetation height was measured during each sampling period.

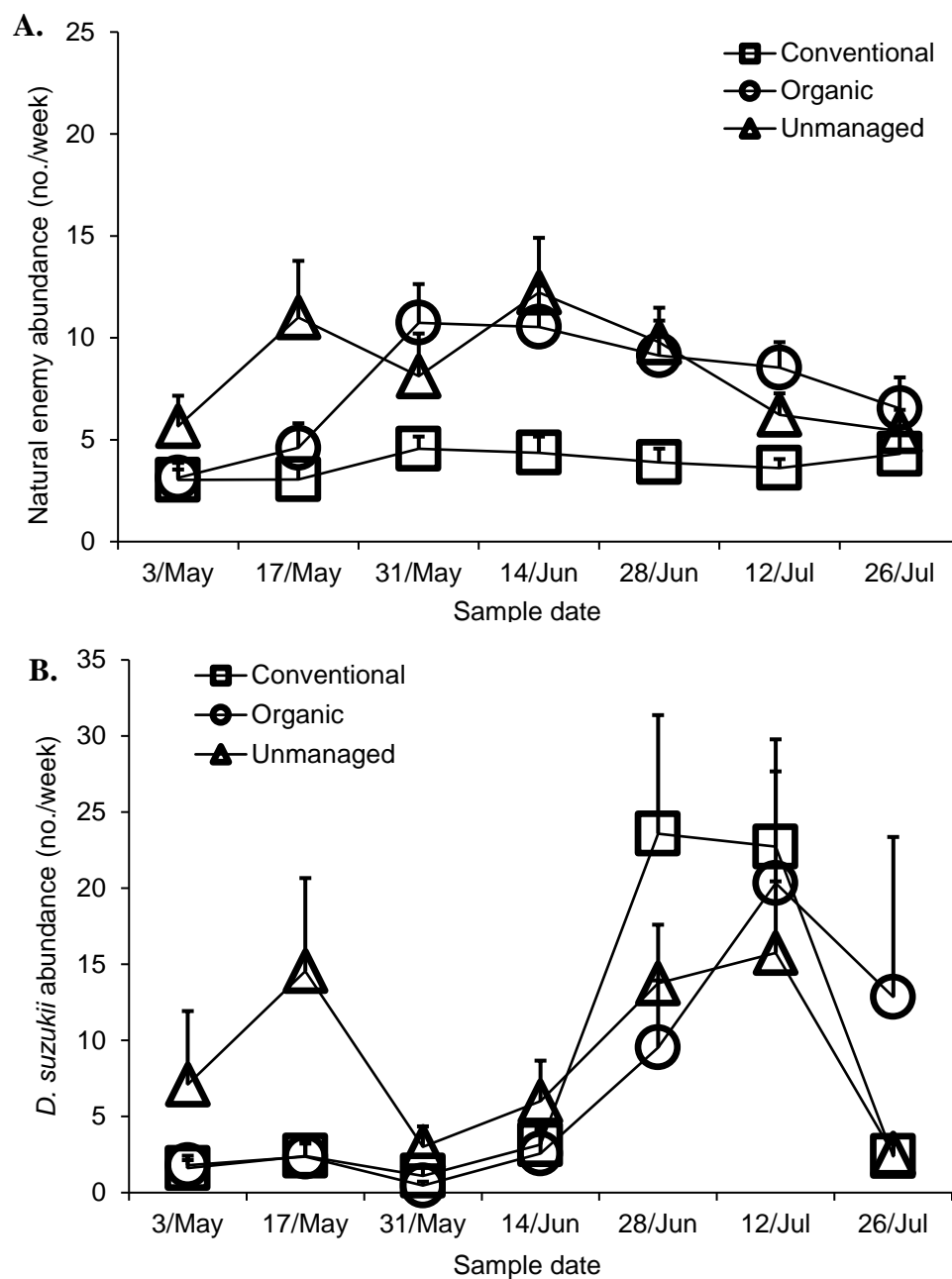
Site	Management practice	Mean vegetation height (m)	Natural enemy abundance	<i>D. suzukii</i> abundance
1	Conventional	0.04	2.24	3.48
2	Conventional	0.09	5.00	0.42
3	Conventional	0.04	2.52	1.60
4	Conventional	0.00	5.95	17.32
5	Conventional	0.00	2.19	1.57
6	Conventional	0.00	3.24	12.58
7	Conventional	0.25	6.38	12.95
8	Conventional	0.09	3.67	16.35
9	Conventional	0.08	3.00	14.80
10	Conventional	0.06	4.52	5.20
11	Conventional	0.37	4.38	3.75
12	Conventional	0.03	2.52	6.00
13	Organic	0.27	7.19	11.43
14	Organic	0.60	10.91	11.65
15	Organic	0.32	9.76	0.95
16	Organic	0.26	4.48	6.32
17	Organic	0.66	5.67	2.95
18	Unmanaged	1.27	7.00	9.86
19	Unmanaged	1.40	6.33	3.90
20	Unmanaged	1.50	12.19	12.10

**Table 3.6.** Landscape factors at all 20 blueberry orchard sites with corresponding arthropod mean abundance during each sampling period. Proportion of non-crop habitat (%) was calculated within a 1 km radius of each orchard site using the 2016 ESRI world imagery dataset. Edge density (ED) and Shannon habitat diversity index (SHDI) was calculated within a 1 km radius of each orchard site using the 2016 USDA cropland data layer to estimate values in a spatial pattern analysis program, FRAGSTATS.

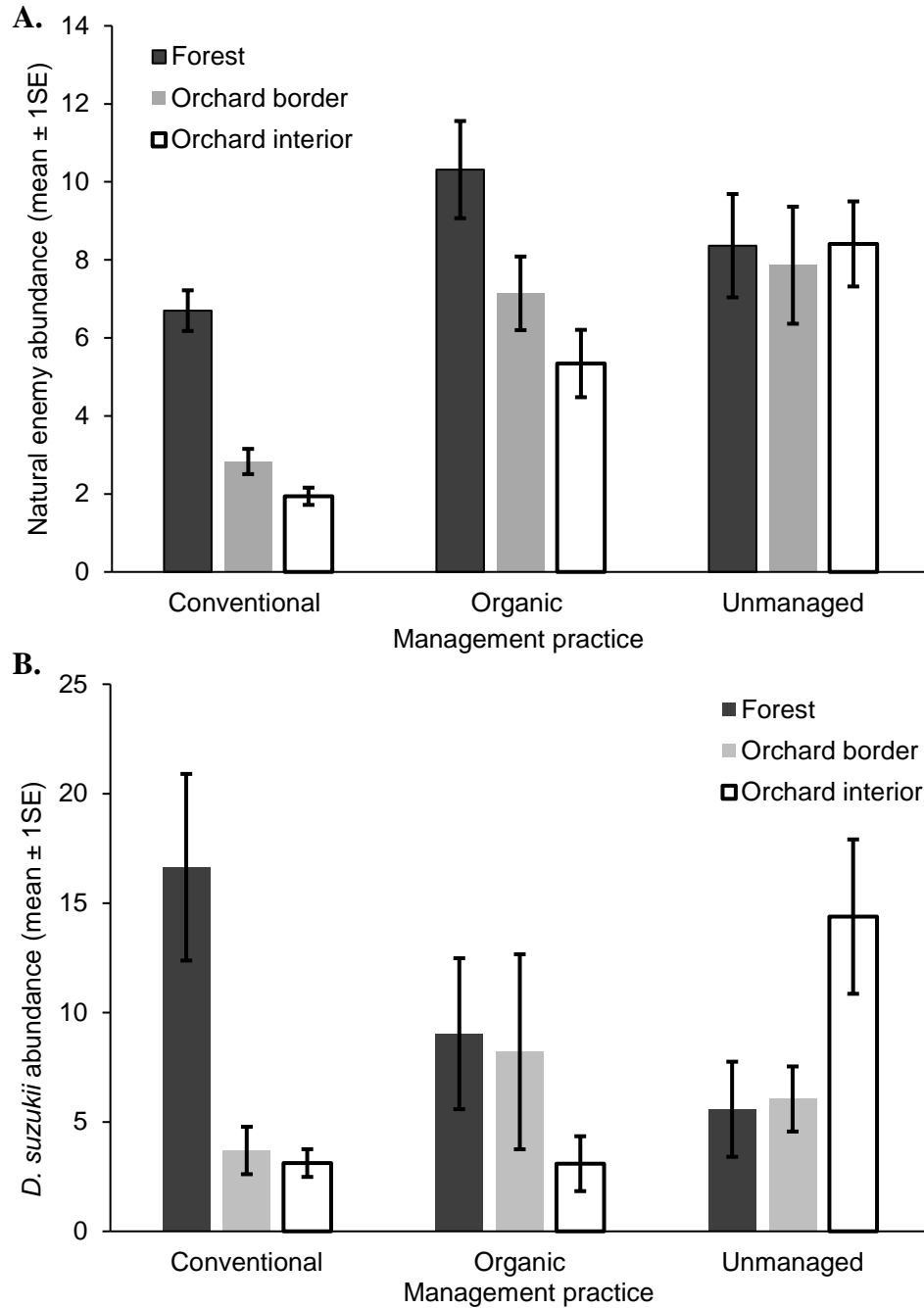
Site	Non-crop (%)	Edge Density	SHDI	Blueberry area (km <sup>2</sup> )	Natural enemy abundance	<i>D. suzukii</i> abundance
1	0.77	253.44	1.81	0.20	2.24	3.48
2	0.39	281.88	2.20	1.19	5.00	0.42
3	0.70	274.09	1.80	0.70	2.52	1.60
4	0.84	337.26	2.17	0.14	5.95	17.32
5	0.98	238.11	1.47	0.07	2.19	1.57
6	0.69	293.87	2.27	0.37	3.24	12.58
7	0.57	283.32	2.14	0.19	6.38	12.95
8	0.71	284.98	1.93	0.18	3.67	16.35
9	0.69	307.94	2.30	0.02	3.00	14.80
10	0.80	194.35	1.74	0.07	4.52	5.20
11	0.54	284.38	2.08	0.81	4.38	3.75
12	0.74	275.38	1.66	0.47	2.52	6.00
13	0.630	280.72	1.77	0.88	7.19	11.43
14	0.48	247.66	2.22	0.38	10.91	11.65
15	0.25	214.52	2.14	0.48	9.76	0.95
16	0.80	283.05	1.89	0.28	4.48	6.32
17	0.59	264.68	2.06	0.60	5.67	2.95
18	0.79	281.77	2.03	0.27	7.00	9.86
19	0.79	289.96	2.05	0.02	6.33	3.90
20	0.84	345.87	2.23	0.14	12.19	12.10



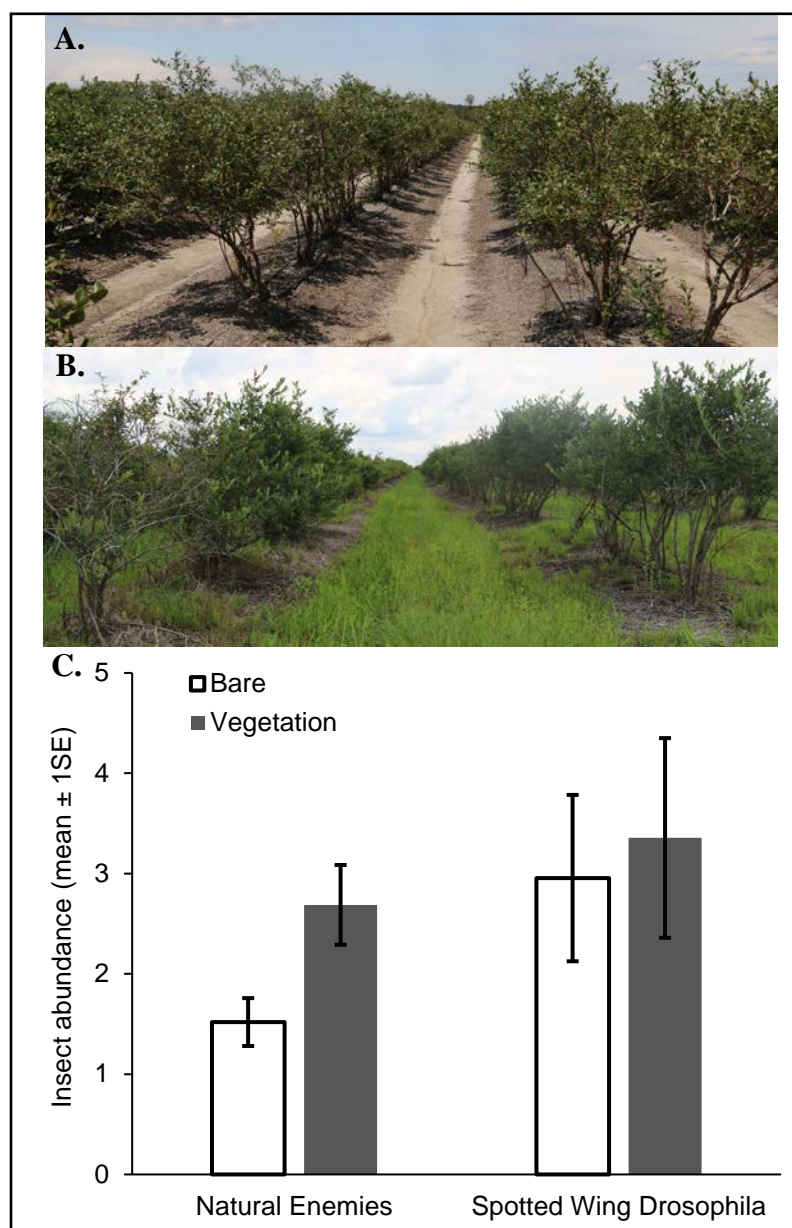
**Fig. 3.1.** The model system located in southeastern United States. A.) Map of study sites located in Georgia with symbols to represent management practice. B.) Example images of a 1 km radius buffer centered at each site using the 2016 USDA cropland data layer and 2016 ESRI world imagery dataset. C.) Sampling design at each orchard along a transect (orchard interior, orchard border, and forest interior) to collect arthropods.



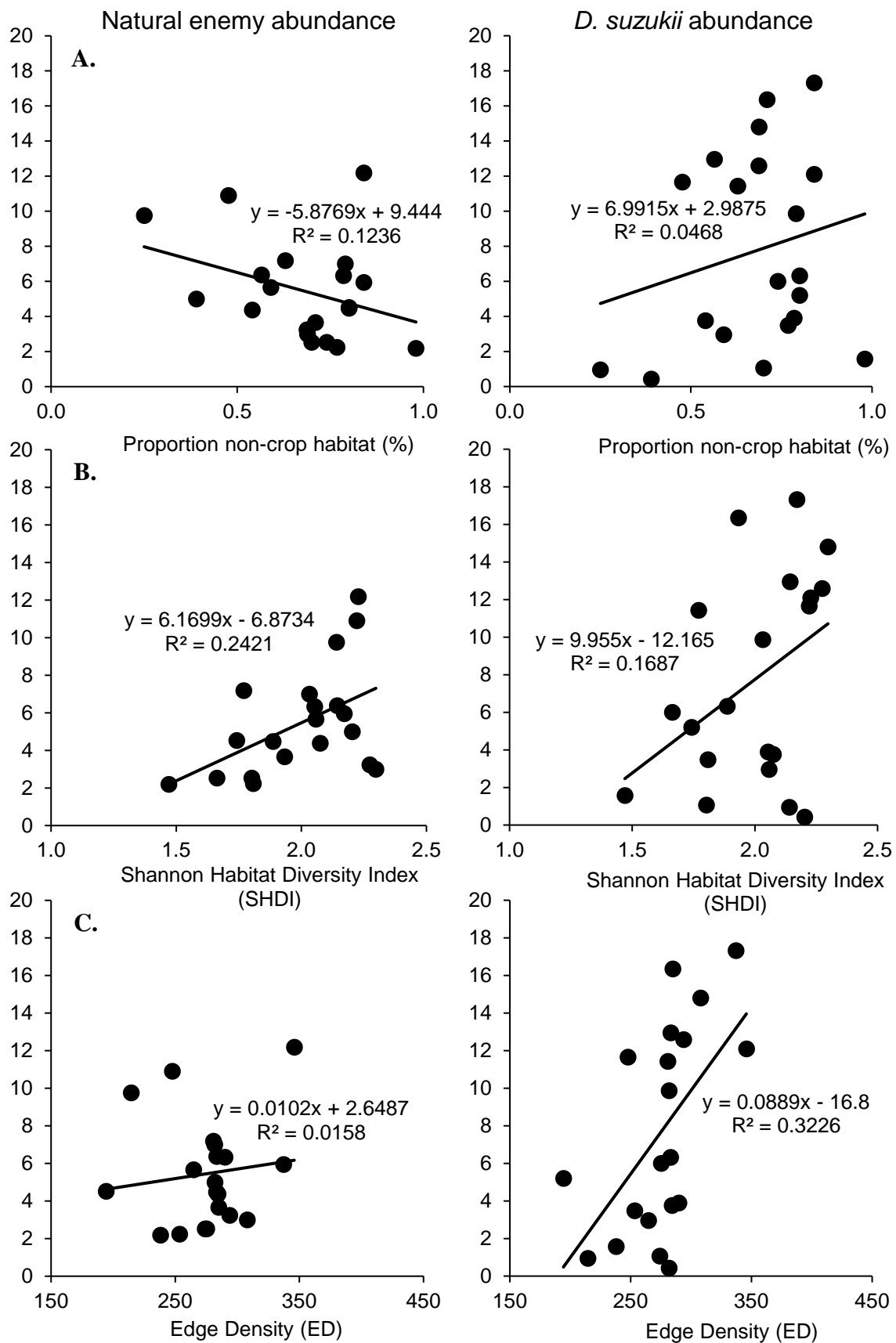
**Fig. 3.2.** Seasonal mean ( $\pm 1$ SE) abundance of A.) natural enemies and B.) *D. suzukii* from suction samples pooled across transect and site to over time. Symbols represent corresponding management practice where samples were collected during the blueberry growing and harvest season.



**Fig. 3.3.** Mean ( $\pm$  1SE) abundance of A.) natural enemies and B.) *D. suzukii* from suction samples pooled across weekly sampling period and site. Bars represent the sampling location (forested habitat, blueberry orchard border, and blueberry orchard interior) in the respective management practice (conventional, organic, and unmanaged).



**Fig. 3.4.** The effect of local vegetation between blueberry crops rows in conventional systems. A.) Represents bare ground between rows and B.) represents vegetation present and commonly mowed. C.) Mean ( $\pm$  1SE) natural enemy abundance and spotted wing drosophila (*D. suzukii*) abundance from suction samples pooled across sampling periods and conventional sites located within the orchard interior. Bars represent the presence of vegetation between blueberry rows.





**Fig. 3.5.** Effects of landscape factors on natural enemy and *D. suzukii* abundance: A.) proportion of non-crop habitat, B.) Shannon habitat diversity index, and C.) edge density. Natural enemy and *D. suzukii* abundances were pooled across all sampling periods and transects for each of the 20 sites. Proportion of non-crop habitat was calculated using a 1 km radius buffer centered at each site measured from the 2016 ESRI world imagery dataset. SHDI and ED were calculated within a 1 km radius buffer centered at each site using the 2016 USDA cropland data layer measured in FRAGSTATS spatial pattern analysis.

## CHAPTER 4

### CONCLUSIONS

The current blueberry agroecosystem investigated is an excellent model system to evaluate the patterns of natural enemy and pest abundances. Blueberry production in the southeastern U.S. is undergoing agricultural intensification, which merits the need for alternative pest control options to alleviate both economic and environmental costs endured. The information provided improves our understanding through the (1) characterization of natural enemies found within the blueberry agroecosystem, (2) the implications of current management practices on natural enemies, (3) the differential effects of the various local and landscape factors on natural enemies and *Drosophila suzukii*, and (4) the potential management tools that may be implemented. Management tools depend on the growing knowledge of how various environmental factors shape arthropods in the region, in which a variety of methods are required in order for growers to improve sustainability and combat pest pressure. Information on the natural enemies in the region, as well as distribution of *D. suzukii* in the environment, help to generate new methods not only for blueberry growers, but landowners interested in improving ecosystem services across the landscape.

The rudimentary finding that conventional management practices negatively impact natural enemy abundance uncovered a plethora of other variables affecting natural enemies in blueberry agroecosystems. Throughout the project, we consistently found a higher abundance of natural enemies distributed towards the adjacent forested habitat in

conventional systems and a more even distribution in the less intensive, unmanaged systems. We could then conclude that a combination of insecticide exposure and availability of non-crop habitat influenced the spatial arrangement of natural enemies within the respective blueberry orchards. Similar distributions were observed by *D. suzukii* abundance where more pests were situated within the forested habitat in managed systems (conventional and organic). Interestingly, there was a higher abundance of *D. suzukii* within the orchard interior of unmanaged systems. This further alludes to the impacts of various environmental factors, leading us to conduct further analysis.

Our results identified the benefit of local vegetation to increase natural enemy abundances, while having neutral effects on *D. suzukii*. This provides support for the implementation of habitat management to incorporate local vegetation without an unwanted increase of *D. suzukii*. The local factors assessed, however, depend on the surrounding landscape. We identified habitat diversity to be the strongest driver in natural enemy abundances, while habitat configuration was the strongest in *D. suzukii* abundance. Between these two taxa, we find differential effects of landscape factors that depend on the mobility and general resource use of natural enemies and *D. suzukii* across the landscape. The response of arthropod abundances helped us to conclude on the relative importance of local and landscape factors found within the studied agroecosystem. Provided with this vital information, we suggest landowners to consider both habitat and landscape management when adopting ecologically-based management practices, which has the potential to enhance ecosystem services provided by natural enemies throughout the landscape.

In summary, this research opens the door towards a more comprehensive pest management system by highlighting the impacts of various environmental factors that shape arthropod abundances. The unique findings within a perennial system located in the southeastern U.S., as well as the comparison of managed and unmanaged orchards, provide insight into the ecological interactions occurring. Future studies will further explore these interactions through the use of molecular gut-content analysis to provide direct feeding linkages between predators and key pests.