ABSTRACT

Title of Thesis: OVERCOMING BARRIERS TO PREDATION: EFFECT OF ALTERNATIVE MANAGEMENT PRACTICES ON PREDATOR-HERBIVORE DYNAMICS IN PRODUCTION NURSERIES

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Alternative pest management tactics were compared to conventional tactics for their affect on pest and beneficial arthropods in production nurseries. Alternatives included habitat manipulations and augmentative releases of predators. Groundcover (weed cloth and hard wood mulch) and container position (pots placed above the ground and pots sunken into the ground [pot-in-pot]) were manipulated in nursery beds. Generally, wood mulch enhanced the abundance of alternative prey and natural enemies at the ground level but not at the soil level within the containers or on plant foliage. The pot-in-pot method strongly increased abundance of both groups at the ground level and the soil level within the containers, whereas pots above ground had a greater abundance of predators on the foliage. Habitat manipulations differentially affected mortality of soil versus foliar herbivores. However, herbivores on the soil in containers suffered greater
mortality than those on the foliage, regardless of groundcover or pot position. Day and
night differences in herbivore mortality and predator taxa were also examined.

Predatory mites were released on *Ilex* at two predator: prey ratios to examine their
ability to suppress *Oligonychus illicis*. Generally, release of predators did not reduce
abundance of *O. illicis* compared to the control. Miticide treated plants had consistently
lower *O. illicis* populations. Leaf damage was less on miticide treated plants. Possible
explanations for the ineffectiveness of augmentative release are discussed.
OVERCOMING BARRIERS TO PREDATION: EFFECT OF ALTERNATIVE MANAGEMENT PRACTICES ON PREDATOR-HERBIVORE DYNAMICS IN PRODUCTION NURSERIES

By

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Thesis submitted to the Faculty of the Graduate School of the University of Maryland, College Park in partial fulfillment of the requirements for the degree of Master of Science 2005

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DEDICATION

I would like to dedicate this thesis to both my lab colleagues and my family. First, to my lab: my fellow lab mates, Steve Frank and Miles Lepping, two people who graciously shared their time and patience with me and unknowingly taught me how to be a better graduate student; Stacey Bealmear and Ellery Vodraska, two wonderful ladies who were always “thrilled” to help with my research day or night and who I am indebted for their efficient skill in the field; and lastly my advisor Paula, who took a chance on an undergraduate long ago, was then stuck with her for two more years, and taught her the joys of research. Secondly, I would like to dedicate this to my family: to Nate, who has been my rock since the very beginning of this research; my sisters, Lisa, Allison, and Sarah, who would make sure that I was doing what I really wanted to do, that I never gave in to pressure from anyone else, and who would offer me food to keep me going; and lastly, to Mom and Dad, who have nurtured and tolerated my interests in insects and science from day one. Their love and support of any of my endeavors and their instillation of creativity and logical thinking has been critical in the past two years and will be essential in the next chapter of my life.
ACKNOWLEDGEMENTS

I would like to thank my other committee members, Galen Dively and Michael Raupp who were always willing to help and advise me through the course of this research and writing process and who were the among the first to always get me to think. I would also like to thank Erin Raupp, the official “transporter” of this thesis. Lastly, I could not have dug so many holes, moved so many plants, and stayed up all night without the help of a few people. I am forever indebted to their field abilities, and their friendship has made long, hot summers full of fun. To Eddie Byrnes, Kim Dickerhoof, Josh Miller, and Sean Skinker, I offer sincere thanks. And words of wisdom that I try to live by: “Try not. Do or do not. There is no try” (Yoda).
TABLE OF CONTENTS

List of Tables ........................................................................................................ vii
List of Figures ........................................................................................................ x

Chapter 1: Habitat Manipulations: Influence of Ground Cover and Pot Position on Natural Enemy Abundance and Herbivore Survival

Introduction ......................................................................................................... 1

Methods

  Study System ................................................................................................. 4
  Experimental Design ....................................................................................... 5
  Arthropod taxa and abundance ...................................................................... 6
  Herbivore Survival ........................................................................................... 7
  Statistical Analysis .......................................................................................... 8

Results

  Arthropod taxa and abundance ..................................................................... 8
  Herbivore survival ......................................................................................... 10

Discussion ......................................................................................................... 10

Chapter 2: Habitat Manipulation: Influence of Management Practices on Spatial and Temporal Predator-Herbivore Dynamics

Introduction ....................................................................................................... 25

Methods

  Study site and experimental design ................................................................. 28

  Arthropod Taxa and Abundance

    Influence of Groundcover and Pot Position .............................................. 30
    Influence of Insecticide on Arthropods ..................................................... 31
Herbivore Survival

Herbivore Survival trials for 2003 ..............................32
Herbivore Survival trials for 2004 ..............................32
Spatial and Temporal Dynamics of Predators ......................34
Statistical Analysis .........................................................34

Results

Arthropod Taxa and Abundance

Influence of Groundcover and Pot Position ......................35
Influence of Insecticide on Arthropods .........................38

Herbivore Survival

Herbivore Survival trials for 2003 ..............................39
Herbivore Survival trials for 2004 ..............................39

Spatial and Temporal Dynamics of Predators ......................41

Discussion .................................................................42

Chapter 3: Augmentative Biological Control: Predatory Mite Releases to Suppress
Southern Red Mite, Oligonychus illicis (Acari: Tetranychidae)

Introduction .............................................................74

Methods

Study System and Experimental Design .........................79
Predatory Mite Treatments ...........................................80
Leaf Damage Ratings ..................................................82
Predatory Mite Lab Feeding Trial ..................................83
Statistical Analysis .....................................................84
Results

Effect of Treatments on *O. illicis* Abundance ..........................84

Leaf Damage Ratings ..................................................................85

Predatory Mite Lab Feeding Trials ........................................85

Discussion ..................................................................................86

References ................................................................................101
LIST OF TABLES

Chapter 1: Habitat Manipulations: Influence of Ground Cover and Pot Position on Natural Enemy Abundance and Herbivore Survival

Table

1  Mean ± SEM abundance of the most common arthropod groups sampled from the ground within wood mulch or weed cloth groundcover habitats summed over four dates in 2002 .................................................................18

2  ANOVA statistics for the abundance of arthropods on the ground within wood mulch or weedcloth groundcover habitats summed over four dates in 2002 ......19

3  Mean ± SEM abundance of arthropod groups sampled from the soil surface within containers of cotoneaster in weed cloth or mulch ground cover plots and either pot-above-ground or pot-in-pot summed over 2 dates (28 August and 13 September 2002) .........................................................20

4  ANOVA statistics for the abundance of arthropods, within containers in weed cloth or mulch groundcover and either pot-above-ground or pot-in-pot containers for summer 2002 .................................................................21

Chapter 2: Habitat Manipulation: Influence of Management Practices on Spatial and Temporal Predator-Herbivore Dynamics

Table

1  Mean ± SEM abundance of the most common arthropod groups sampled from the ground within wood mulch and weed cloth groundcover habitats for 6 August 2003 .................................................................53

2  a. ANOVA statistics for the abundance of arthropods within wood mulch or weed cloth groundcover habitats for 6 August 2003.
   b. Statistics for the abundance of arthropods within wood mulch or weed cloth groundcover habitats for 6 August 2003. Analysis was done using the Sheirer-Ray-Hare non-parametric method ................................................................. 54

3  Mean abundance (± SEM) of the most common arthropod groups sampled from the ground within wood mulch and weed cloth groundcover habitats summed over 3 dates in 2004 .................................................................55

4  Mean ± SEM abundance of the most common arthropod groups sampled
from the ground within wood mulch and weed cloth groundcover habitats summed over 3 dates in 2004………………………………………………………………………………..56

5 Mean ± SEM abundance of the most common arthropod groups sampled from the soil surface within pot-above-ground and pot-in-pot plant containers in wood mulch and weed cloth groundcover habitats for 6 August 2003……………………………………………………………………………….57

6 a. ANOVA statistics for the abundance of arthropods sampled from the soil surface within containers from both wood mulch and weed cloth groundcover habitats and pot-in-pot and pot-above-ground containers for 6 August 2003. b. Statistics for the abundance of arthropods within wood mulch or weed cloth groundcover habitats for 6 August 2003. Analysis was done using the Sheirer-Ray-Hare non-parametric method………………………………………………58

7 Mean ± SEM abundance of the most common arthropod guilds sampled from the soil surface within plant containers in wood mulch and weed cloth groundcover habitats and pot-above-ground and pot-in-pot containers summed over 3 dates in 2004……………………………………………………………………………….59

8 ANOVA statistics for the abundance of arthropods sampled from the soil surface within containers from both wood mulch and weed cloth groundcover habitats and pot-in-pot and pot-above-ground containers for summer 2004…………………………………………………………………………60

9 Mean ± SEM abundance of the most common arthropod groups sampled from ground pitfall traps in plots treated with insecticide and not treated summed over 3 dates in 2004……………………………………………..61

10 ANOVA statistics for the abundance of arthropods sampled from ground pitfall traps in plots treated and untreated with insecticide for summer 2004…..62

11 Mean ± SEM abundance of the most common arthropod groups sampled from within plant containers in plots treated and untreated with insecticide for 3 dates in summer 2004………………………………………………………………63

12 ANOVA statistics for the abundance of arthropods sampled from within containers from both treated (Insecticide) with or untreated (No Insecticide) for 3 dates in summer 2004……………………………………………………64

13 Chi – square statistics for the survival of S. exigua comparing the three main treatments of groundcover, pot position, and herbivore location during 3 separate field trials in summer 2003……………………………………………………65

Table

1. ANOVA contrasts for evaluating *Oligonychus illicis* egg and active stage abundance singularly and combined for *O. illicis* eggs and active stages for the control, miticide, and predatory mite (both 1:25 and 1:50) treatments……95

2. Mean ± SEM of predatory mites (per cm² leaf area) recovered during sampling for the abundance of *Oligonychus illicis* eggs and active stages on three dates in 2004: 7 July, 4 August, and 26 September…………………………96
LIST OF FIGURES

Chapter 1: Habitat Manipulations: Influence of Ground Cover and Pot Position on Natural Enemy Abundance and Herbivore Survival

Figure

1  a. Layout of treatment plots with weed cloth and wood mulch groundcover, and pot-above-ground and pot-in-pot, and placement of pitfall traps used for sampling (represents one replicate). 1b. Diagram of pot-in-pot and pot-above-ground treatments................................................................. 22

2  Mean proportion (SE) of Corythuca cydoniae (Hawthorn lace bug, HLB) nymphs remaining 2 days after plants were infested. Containerized plants were in beds with wood mulch and weed cloth groundcover. Within each groundcover treatment, containers were above ground or pot-in-pot. The study was repeated on three dates: a. 24 June 2002, b: 9 July 2002, and c: 5 August 2002………………..23

3  Mean (± SEM) number of O. sulcatus (black vine weevil, BVW) eggs remaining 1 day after 10 eggs were placed on a petri dish at the soil surface in containers of Cotoneaster dammeri. Containerized plants were in beds with wood mulch and weed cloth groundcover; within each groundcover treatment, containers were above ground or pot-in-pot. The study was repeated on three dates: 12, 13, 14 August 2002. There was no significant effect of date, therefore, the figure represents the pooled values for the three dates……………………………………24

Chapter 2: Habitat Manipulation: Influence of Management Practices on Spatial and Temporal Predator-Herbivore Dynamics

Figure

1  Layout of treatment plot (either weed cloth or wood mulch groundcover) and pitfall traps.................................................................66

2  Mean abundance of spiders pooled (a) and the spider families: Salticidae (b), Araneidae (c), and Thomisidae (d) sampled from foliage by beating during the day in September 2004.................................................................67

3  Mean abundance of spiders pooled (a) and the spider families: Salticidae (b), Thomisidae (c), and Anyphaenidae (d) by beating azalea foliage during the night in October 2004.................................................................68

4  Stephanitis pyrioides azalea lace bug (ALB) survival over the season in 2004 with two groundcover types (wood mulch and weed cloth) and two positions (pot-in-pot and pot-above-ground)........................................69
5 Mean proportion of *Agrotis ipsilon*, black cutworm (BCW) mortality for day (a) and night (b) in September 2004. Values are pooled across ground covers of wood mulch and weed cloth.............................70

6 The mean number of observed predation events during corresponding *Agrotis ipsilon* survival trials during the day (a) and at night (b). Predation event frequencies are total number of predation events observed over the length of each trial. Values are pooled across ground cover.........................71

7 The mean number of acts of predation observed by predator family for both day (a) and night (b) that correspond to the *Agrotis ipsilon* survival trials. Values are summed over the course of the respective trial............................72

8 Mean number of arthropod families observed during the day and night *Agrotis ipsilon* survival trials consuming larvae in September 2004........73

Chapter 3: Augmentative Biological Control: Predatory Mite Releases to Suppress Southern Red Mite (*Oligonychus illicis*) (Acari: Tetranychidae)

Figure

1 The abundance (number per cm² leaf area) of *Oligonychus illicis* eggs alone (a), active stages alone (b), and eggs + active stages (c) with the fourthreatments: control, miticide, and predator: prey ratios of 1:25 and 1:50. The 28 August 03 is a precount of *O. illicis* abundance prior to treatments. Arrows with letters indicate: (a) predator and miticide application (Hexygon), (b) predator application, and (c) predator and oil application. Note differences in scale……97

2 Leaf damage ratings cause by *Oligonychus illicis* for *Ilex* leaves collected on 26 August 2004. Ratings equal to 1 are 1 to 5% damage, 2 are 6 to 10% damage, and 3 are 11 to 15% damage…………………………………..98

3 A “no choice” lab predator preference study measuring the percentage survival of either eggs or active stages of *Oligonychus illicis* when they were offered singularly to two predatory mite species, *Galendromus occidentalis* (GO) and *Neoseiulus fallacis* (NF)……………………………….. 99

4 A “choice” lab predator preference study to measure percentage of survival of *Oligonychus illicis* eggs and active stages, when two predators, *Galendromus occidentalis* (GO) and *Neoseiulus fallacis* (NF), were offered both life stages in the same microcosm……………………………………100
Chapter 1

Habitat Manipulations: Influence of Ground Cover and Pot Position on Natural Enemy Abundance and Herbivore Survival

INTRODUCTION

Managed ecosystems undergo frequent disturbance that often render them unfavorable for natural enemies (Van Driesche and Bellows 1996, Landis et al. 2000). For example, the process of urbanization or the production of agricultural crops alters the vegetational diversity and structural complexity of a habitat reducing the availability of resources necessary for natural enemy survival and reproduction (Winchester 1997, Rypstra et al. 1999, Landis et al. 2000). Other plant or crop management practices, such as the application of insecticides (Raupp et al. 1992, Landis et al. 2000, Lee et al. 2001) or field tillage (Rypstra et al. 1999, Halaj et al. 2000), also disturb the arthropod community composition and detrimentally affect natural enemies. These disturbances disrupt functional processes governing herbivore - natural enemy dynamics and often result in more frequent herbivore outbreaks (Roland and Taylor 1997, Landis et al. 2000). Therefore, there is a need to identify measures to restore functional processes driving herbivore – natural enemy dynamics and reduce the likelihood of pest outbreaks in managed ecosystems.

Habitat manipulation, a form of conservation biological control, specifically alters habitats to improve the availability of resources required by natural enemies for their optimal performance (Landis et al. 2000). Habitats that provide: alternative food sources such as nectar and pollen for natural enemies; shelter in which natural enemies can overwinter and seek refuge from factors such as environmental extremes, pesticides, or
other natural enemies, and alternative hosts or prey should favor natural enemies (Landis et al. 2000). Therefore, it should be possible to manipulate managed habitats to favor natural enemies and enhance their populations, and reduce pest insect outbreaks (Gurr et al. 2000).

Numerous studies have examined the influence of the vegetational diversity and structural complexity of a habitat on herbivore and natural enemy dynamics and the manipulation of these variables for their potential use as conservation biological control practices. Most of these studies have been conducted in traditional agricultural systems with fewer studies in orchards and production nurseries and urban ornamental systems. Variables known to effect herbivore and/or natural enemy dynamics include floral resources (White et al. 1995, Patt et al. 1997, Shrewsbury et al. 2004, Rebek et al. 2004); hedgerows (Rieux et al. 1999, Landis et al. 2000, Lee et al. 2001); polycultures, intercropping, and cover crops (Cárcamo 1994, Rypstra et al. 1999, Landis et al. 2000); tillage (Rypstra et al. 1999, Halaj et al. 2000, Symondson et al. 2002); structural complexity (Rypstra et al. 1999, Uetz 1999, Shrewsbury et al. 2004, Shrewsbury in press); and mulch (Riechert and Bishop 1990, Brust 1994, Rypstra et al. 1999, Halaj et al. 2000, Johnson et al. 2004). Most of these studies found habitats with greater vegetational diversity and/or structural complexity had fewer pest outbreaks compared to simpler habitats (see reviews by Gurr et al. 2000, Landis et al. 2000, Langellotto et al. 2004). Additionally, many studies found a greater abundance of natural enemies in complex habitats compared to simpler ones (Gurr et al. 2000, Landis et al. 2000, Symondson et al. 2002, Shrewsbury in press). Few studies have identified the mechanisms underlying
relationships between vegetational diversity / structural complexity, herbivores, and their natural enemies (Langellotto and Denno 2004, Shrewsbury in press).

In addition to influencing the availability of resources for natural enemies, management practices may also result in physical barriers that reduce the accessibility of herbivores to natural enemies and ultimately the ability of natural enemies to reduce herbivore populations. For example, the addition of barriers limited access of ground dwelling predators to plant material (Clark et al. 1994, Päts et al. 1999, Vichitbandha et al. 2002). Clark et al. (1994) found that in constructing artificial fences of galvanized steel strips carabid and spider were unable to gain access to field plots. In addition, plant damage was greater in plots where barriers excluded ground predators than plots where plants remained accessible to natural enemies (Clark et al. 1994).

Two conventional practices in ornamental nurseries that likely influence the accessibility and availability of herbivores to natural enemies are the use of plastic containers and black weed cloth, a substrate where containers with plants are lined out on top. Weed cloth may provide a harsh, barren environment with little refuge or food for predators. This is likely an unfavorable environment, especially for ground dwelling predators to cross to access plants and herbivores. Plastic containers are smooth and slippery and may be difficult for certain predators to climb resulting in a physical barrier between ground dwelling predators and herbivores on plants growing in containers.

Alternative management practices to weed cloth and above ground container placement are hard wood mulch as a ground cover and containers sunken into the ground (pot-in-pot planting system). Wood mulch was examined because previous research has shown that mulch can enhance the abundance of generalist predators in agro-ecosystems
by creating more favorable habitat (Reichert et al. 1990, Brust 1994, Halaj et al. 2000, Jordan et al. 2002). The pot-in-pot method is a relatively new production method and its adoption by container plant producers is increasing (Hall et al. 2002). In this system, an empty plant container is placed in the soil, and a container with a plant is then placed in this “socket pot” with the lip of the containers at soil level. This method was examined because it should make plants and herbivores more accessible to ground predators. However, no one has examined the influence of the above ground or pot-in-pot method on the ability of predators to access herbivores on the plants.

The overall objective of the study was to identify habitat manipulations that enhance predator populations and reduce pest insect outbreaks in ornamental nursery systems. The specific objectives were to: 1) determine the effects of a groundcover tactic (wood mulch and weed cloth) on herbivore survival and arthropod taxa and abundance, and 2) determine the effect of pot position (pot-in-pot or pot-above-ground) on herbivore survival and arthropod taxa and abundance.

MATERIALS AND METHODS

Study System. Field studies were conducted from June to September 2002 at the University of Maryland, Upper Marlboro Research and Education Facility in Upper Marlboro, Maryland. Two treatment plots were arranged in five blocks (replicates), and each plot simulated ornamental container plant production beds. Each treatment plot was 6.3 m x 7 m in size and separated by 10.5 m turf buffer within a replicate. Replicate blocks were separated by a distance of at least 20 m. All plots were 8.75 m from a wood edge. *Cotoneaster dammeri* var. ‘Royal Beauty’ were used as study plants. Cotoneasters are broad leafed evergreen woody shrubs commonly grown in container production.
After plants were purchased, they were transplanted from 3.79 L to 11.34 L containers with Pro-Mix (Scotts-Sierra Horticultural Products Company, Ohio) sterile potting medium. In each treatment plot, plants were placed in three rows with 6 plants per row (18 plants within each plot). Containers were 1 m apart within and between rows (Fig. 1a). Plants were watered as needed (~3 times per week) throughout the growing season. All plots were hand weeded, and grassy areas directly adjacent to the plots were mowed periodically to maintain a low turf/weed cover.

Two herbivores were used as sentinel prey in this study, Hawthorn lace bug, *Corythuca cydoniae* Fitch (Heteroptera: Tingidae) and black vine weevil, *Otiorhynchus sulcatus* Fabricus (Coleoptera: Curculionidae). Both herbivores are key pests in nurseries. *Corythuca cydoniae* is a foliar pest of cotoneaster. The damage caused by feeding on the leaves is characterized by stippling (small yellow to white leaf spots caused by the removal of chlorophyll). *Otiorhynchus sulcatus* is a pest of roots, as larvae and foliage, as adults, of many herbaceous perennials and woody shrubs. For field trials, I used the egg stage, which occurs in the upper soil surface. Using these two herbivores provided knowledge of the effect of the treatments on foliar and soil dwelling pests.

**Experimental Design.** The experiment was a 2 x 2 factorial split-plot design blocked by location with 1 replicate per block (=5 blocks). Within each replication, there were two treatment levels of groundcover (whole plot): hard wood mulch and weed cloth. Within each groundcover treatment, there were two treatment levels of planting method (subplot): pot-above-ground and pot-in-pot. One of two plots in each replicate was randomly assigned to a groundcover treatment. The mulch that covered the designated mulch plot was hardwood mulch (average chip size 2 x 3 cm) and spread across the plot.
cm deep; this mulch layer covered a sheet of weed cloth to deter weed growth (as growers would use). The other plot was covered with black weed cloth (Polypropylene, SI Performance Technology, Ringgold, GA). Pot-in-pot and pot-above-ground treatments were randomly assigned to half of the containers in each groundcover treatment plot. Therefore, nine plants were pot-in-pot and nine plants were pot-above-ground (Fig. 1a).

**Arthropod taxa and abundance.**

*Activity within the plot.* Sampling was conducted by placing 3 pitfall traps in each of the study plots. Traps were evenly spaced in 1 of 3 rows within each plot, and the row of traps was placed 2.74 m from the long side of the plot (Fig. 1a). Traps were plastic cups (Solo Cup Company, Urbana, Illinois), with a diameter of 9 cm, that were submerged into the ground with the lip of the cup even with the soil surface. To prevent entry of rain or irrigation water into the traps, a cover was made with plastic plate cover supported by three 15 cm carriage bolts. The covers were placed 5 cm above the traps to allow arthropods to freely move under the covers. All traps were filled with approximately 150 ml of 1:5 propylene glycol: water solution. Sampling occurred on 17 July, 14 August, 28 August, and 13 September 2002. Traps remained in the plot for one week intervals. Samples were collected, returned to the laboratory, rinsed, and stored in 70% ethyl alcohol. At a later date arthropods were sorted and identified to family level and trophic group (alternative prey or natural enemies). Potential alternative prey consisted of all arthropods (as in Halaj et al. 1998, Shrewsbury in press) except *C. cydoniae*. We used a liberal estimate of alternative prey in the absence of dietary
information for the predators encountered (Halaj et al. 1998, Shrewsbury in press). The natural enemy guild consisted of strict predators, omnivores, and parasitoids.

Activity within plant containers. Sampling was conducted by placing pitfall traps inside the containers. Pitfall traps were as described above except traps were filled with 75 ml of propylene glycol solution and placed in one randomly selected pot-in-pot and pot-above-ground container in each of the groundcover treatment plots. These cups were submerged so that the cup lip was even with the soil level within the pot. Sampling was conducted on 28 August and 13 September 2002; these dates coincided with herbivore survival trials (see next section). For each sampling date, cups remained in each pot for one week at which time samples were collected, stored, and sorted as above.

Herbivore survival.

*Corythuca cydoniae* survival was measured. Two cotoneaster plants (subsamples) on each half of every plot were selected at random and infested with *C. cydoniae* nymphs so that there were 4 plants total infested per groundcover treatment (2 plants in above ground and 2 plants in pot in pot treatments). Three trials were conducted throughout the summer. The number and life stage of *C. cydoniae* varied per trial due to limited availability of *C. cydoniae* from field populations. The first trial was conducted on 24-25 June 2002, and each study plant was infested with 30 2\textsuperscript{nd} or 3\textsuperscript{rd} *C. cydoniae* nymphs, with 10 lace bugs placed on each of 3 branches. The second trial was conducted on 9 July 2002 and each study plant was infested with 15 5\textsuperscript{th} instars per plant, with 5 lace bugs nymphs placed on each of 3 branches. The final trial on 5 August 2002 used 20 3\textsuperscript{rd} or 4\textsuperscript{th} instars, with either 6 or 7 lace bug nymphs on each of three branches. Post counts of live lace bugs were taken 2 days after plants were infested to determine *C. cydoniae* survival.
Survival studies using *O. sulcatus* eggs were also conducted. Three trials were conducted on August 12, 13, and 14, 2002. Lids of small petri dishes (6 cm diameter) were lightly brushed with egg white to aid adhesion, and 10 *O. sulcatus* eggs were placed on the petri dish. Petri dishes were placed on the soil surface so that the petri dish rim was at soil level in one plant container from each pot position treatment within the mulch and weed cloth ground cover treatments. The number of eggs remaining was determined one day after eggs were placed in the pots.

**Statistical Analysis.** An analysis of variance (ANOVA) for a split plot factorial design (Proc Mixed, SAS Institute 1999) was used to determine the effect of ground cover and pot position, and any interaction between the two treatments on abundance of arthropod taxa and trophic groups within the study plot and within plant containers, and *C. cydoniae* and *O. sulcatus* egg survival. Arthropod data from pitfall traps were pooled across sampling dates. *Otiorhynchus sulcatus* egg survival was also pooled across 3 dates. To determine if data met the assumptions of ANOVA, homogeneity of variances and normality were examined using Proc Univariate and plotting residuals (SAS Institute 1999). Data that did not meet the assumptions of normality and homogenous variances were log_{10} transformed before analysis was performed on transformed data. Data are presented as untransformed means ± SEM. All analysis incorporated a blocking term. Differences were considered significant at \( P < 0.05 \).

**RESULTS**

**Arthropod taxa and abundance.**

*Activity within the plot.* Collembola, a potential alternative prey item, were more abundant in wood mulch plots than in weed cloth plots (Tables 1 and 2). There were
several abundant families within the natural enemy group. Araneae and Formicidae abundance did not significantly differ between wood mulch and weed cloth (Tables 1 and 2). The most abundant family of spiders, Lycosidae, also did not significantly differ between the two habitats, but lycosids were numerically greater in weed cloth plots (Tables 1 and 2). However, Staphylinidae and Carabidae were significantly more abundant in the mulch plots (Tables 1 and 2). Other abundant taxa collected were Scelionidae, hymenopteran parasitoids, and Gryllidae, field crickets. Scelionidae and Gryllidae did not significantly differ in abundance when mulch was present or absent (Tables 1 and 2). However, Gryllidae abundance was numerically greater in the plots with weed cloth.

*Activity within plant containers.* There was no significant interaction between groundcover and pot position and no significant effect of groundcover on arthropod abundance, though abundance was generally greater in the presence of mulch (Table 3).

The most abundant taxon within alternative prey was Collembola. There was a significant interaction of groundcover and pot position on Collembola abundance (Tables 3 and 4) where more Collembola were collected in traps from pot-in-pot containers in wood mulch plots. There were several abundant taxa within the natural enemy group. Two ground dwelling predator families, Staphylinidae and Carabidae, were significantly more abundant in pot-in-pot containers (Tables 3 and 4). Two other groups, Formicidae and Gryllidae, were also significantly more abundant in the pot-in-pot containers than in the pot-above-ground (Tables 3 and 4). Spiders (Araneae) and Scelionidae, hymenopteran parasitoids, showed a significant mulch by pot position interaction (Table
3 and 4); more spiders and Scelionids were sampled in the mulch, pot-in-pot containers than in the mulch, above ground or weed cloth treatments.

**Herbivore survival.**

In all three experiments examining *C. cydoniae* survival, there was no significant interaction between groundcover and pot position treatments (24 June 2002: $F= 0.00$, df $= 1, 32, P= 0.9790$; 9 July 2002: $F= 0.34$, df $= 1, 28, P = 0.5642$; 5 Aug. 2002: $F = 0.74$, df $= 1, 36, P = 0.3951$) (Fig. 2). For all survival studies, there was no significant main effect of groundcover on *C. cydoniae* survival (26 June 2002: $F=0.03$, df$= 1, 32, P= 0.8541$; 9 July 2002: $F=0.21$, df$=1, 8, P=0.6573$; and 5 Aug 2002: $F=2.21$, df$= 1, 36, P=0.1458$). The survival study on 9 July 2002 did show a significant effect of pot position ($F=6.76$, df $=1, 28, P=0.0147$) on *C. cydoniae* survival, with lower survival of *C. cydoniae* in the pot-in-pot containers (Fig. 2).

There was no significant effect of date ($F= 0.10$, df$= 2, 43.2, P= 0.9048$) on *O. sulcatus* egg survival so data were pooled across three dates. There was no significant interaction between ground cover and pot position ($F= 0.07$, df$= 1, 55, P= 0.7901$) or main effect of ground cover ($F= 3.03$ df$= 1, 55, P= 0.0872$) on *O. sulcatus* egg survival. However, there was significantly fewer BVW eggs remaining in the pot-in-pot containers than in the pot-above-ground containers ($F= 79.97$, df$= 1, 55, P < 0.0001$) (Fig. 3).

**DISCUSSION**

Overall, the type of groundcover used in the simulated nursery beds influenced the abundance of alternative prey and natural enemy taxa active within the plot. However, this effect varied with arthropod taxa. Moreover, containerized plants in the pot-in-pot system consistently had a greater abundance of natural enemies than containers
in the above ground system. For some taxa, such as Collembola and spiders, this effect was enhanced in mulched plots. Surprisingly, the increase in natural enemy activity did not consistently transfer to greater predation of herbivores.

Collembola also had a greater abundance in wood mulch plots. Environmental factors that influence Collembola distribution include moisture (Frampton et al. 2000), food, soil type, and population density (Bengtsson et al. 1994). Although untested in this study, the presence of wood mulch could have influenced these factors to favor Collembola, whereas weed cloth would not. Mulch and the limited shade provided by the plants may have increased moisture levels, as reported by Porter (1998). Badejo et al. (1998) also showed that soil moisture was positively correlated with populations of soil Collembola in an agroforestry system when trees were mulched with tree prunings. The quality and quantity of the litter also affected soil Collembolans (Badejo 1998). Slowly decomposing and larger quantities of tree litter supported more Collembola because there were more favorable microenvironments as well as a larger food supply and shelter (Badejo 1998). It is likely that Collembola emigrated into my wood mulch plots from surrounding turf and crop areas. Higher levels of moisture and available food resources enhanced collembolan reproduction in wood mulch plots.

There was a greater abundance of natural enemy families in wood mulch plots, such as staphylinids and carabids, than weed cloth plots. Other studies have shown that Collembola are prey for carabids (Bauer 1982, 1985, Bilde et al. 2000), staphylinids (Bauer and Pfeiffer 1991), and spiders (Sunderland et al. 1986). The distribution and abundance of Collembola may have influenced the abundance of predators in the plots. Similarly, other studies have found alternative prey (Settle et al. 1996, Symondson et al.
2002, Shrewsbury in press), including Collembola (Potts and Vickerman 1974), to be a potential mechanism influencing natural enemy abundance. Settle et al. (1996) manipulated the organic matter in rice paddies plots, enhanced the population of detritus and plankton-feeding insects, and ultimately increased the abundance of generalist predators. Pest populations in these plots were maintained at lower levels than plots that received an early season insecticide treatment (Settle et al. 1996). Shrewsbury (in press) reported that the dominant predator in an azalea (Rhododendron sp.) system was a spider, Anyphaena celer (Araneae: Anyphaenidae). Its abundance was highest on plants that had greater densities of alternative prey (Cicadellidae).

Previous studies utilizing a mulch groundcover have had variable success in attracting both carabid and staphylinid beetles. Similar to my study, Halaj et al. (2000) and Johnson et al. (2004) reported an increase in beetle abundance with straw mulch refugia or straw mulch groundcover, respectively. Whereas, Prasad and Henderson (2002) reported a more variable straw mulch effect on the abundance of carabids compared to staphylinids. Ground beetles comprised a smaller proportion of total natural enemies in straw plots compared to control plots, and this proportion gradually decreased through the growing season. However, staphylinids had a larger abundance in mulched than control plots. The authors concluded that the two beetles respond differently to microenvironmental changes. In addition, Braman et al. (2000) found that there abundance of carabids and staphylinids were not enhanced in wood chip mulched beds. More carabids and staphylinids were collected from pitfall traps in turf plots instead.

In my study, there was no significant difference in spider abundance in plots with or without mulch. This is contrary to the results of Reichert and Bishop (1990), Halaj et
al. (2000) and Johnson et al. (2004) that all showed significant increases in spider abundance in mulch treatments, particularly the spider families Lycosidae and Linyphiidae (Reichert and Bishop 1990, Johnson et al. 2004). The most common spider family in my study was wolf spiders (Lycosidae), followed by jumping spiders (Araneae: Salticidae) and crab spiders (Araneae: Thomiscidae). Wolf spiders did not show a significant difference in abundance ($P = 0.09$) in the two groundcover types, but there was a numerical trend for higher abundance in the weed cloth plots. Differences in the response of spiders to mulch in this study and those of other published studies might be due to the type of mulch used. Alternatively, the greater abundance of lycosids in weed cloth compared to mulch plots may be due to an aggregative response of lycosids to a potential prey item, such as field crickets (Maloney et al. 2003). In my study, wolf spiders also had a numerical dominance in weed cloth plots where field crickets were very abundant. Persons and Uetz (1996) showed that wolf spiders might use chemical cues as a source of information while foraging. *Schizocosa ocreata* (Hentz 1844), a wolf spider common to forested areas of the Northeast, spent significantly longer periods of time on substrata where crickets had walked upon. However, this was a laboratory experiment, where external features, such temperature, moisture, and predation, were controlled. The authors were uncertain to what degree spiders use chemical cues in a natural setting.

A third player in this cricket – wolf spider complex are the parasitoids in the family Scelionidae that was also very abundant in the mulch and weed cloth study plots. Previous work by Masner (1978) found that scelionids in the genus *Leptoteleia* Kieffer are egg parasitoids of crickets. Crickets lay their eggs in firm bare, soil from late July
through September; eggs are the overwintering life stage (Merchant 2001). The high abundance of scelionids in the study plots could be the result of high host availability, especially in weed cloth plots.

Ants (Hymenoptera: Formicidae) comprised one of the largest families of insects captured, though abundance did not appear to be affected by groundcover type. In other systems, ants are sometimes included as predators (Cockfield and Potter 1984, Smitley et al. 1998, Kunkel et al. 1999). In addition, certain species of ants are known to be herbivores, fungivores, and honeydew feeders (Folgarait et al. 1996, Davidson et al. 2003, Hunt 2003, Mathews 2004, Maier and Potter 2005). However, the role that ants played in this study is uncertain. As ant abundance did not differ between groundcover types, it appears that ants were foraging in habitats regardless of habitat differences. Ants were frequently observed foraging in the canopy of *C. dammeri*, but predation activity was not witnessed during sampling. It is important to elucidate, in the future, the roles of ants as potential predators.

Interestingly, the positive response by staphylinids and carabids to the addition of mulch at the plot level did not transfer to any significant interactive effect between mulch and pot position. Staphylinids and carabids were more abundant in containers in the pot-in-pot planting system, regardless of the groundcover type present in the plot.

On the other hand, Araneae did not differ in abundance at the ground level between plots with mulch and weed cloth. However, when mulch was present, more spiders moved into containers in the pot-in-pot system, then containers in the same system with weed cloth. It is likely that spiders in this system were not good climbers of the plastic containers. When this barrier was removed (e.g. pot-in-pot system), it appears
spiders responded aggregatively to alternative prey abundance as in Harwood et al. 2001, or specific spider taxa shared a similar preference with alternative prey for a common microhabitat (Bonte and Mertens 2003).

We found variable effects of the habitat manipulations on foliar and ground herbivores. For the foliar herbivore, *C. cydoniae*, groundcover type had no effect on survival while pot position had a variable effect with lower survival in the pot-in-pot containers in one of three trials. In contrast, survival of the ground herbivore, *O. sulcatus* eggs, was slightly lower (*P = 0.09*) in mulch than weed cloth. Moreover, there was very low survival in containers in the pot-in-pot system, regardless of groundcover type. It appears predators that are attracted to mulch and forage within plant containers may not climb into *C. dammeri* canopies, by choice, or they may not possess the ability to do so. Predators’ inability to climb or their preference not to climb resulted in lower mortality of *O. sulcatus* on the soil of pot-above-ground containers. Therefore, it is assumed that predators were attracted to the mulched habitat, foraged within the containers as the result of easier access, and consumed *O. sulcatus* eggs. Staphylinids and carabids were the only predator taxa to increase in mulch plots. The majority of individuals within these taxa are ground foraging (Triplehorn and Johnson 2005). Also, more of these predators were trapped in pot-in-pot containers. Staphylinids and carabids were either more effective ground than foliar foragers (Wyman et al. 1976) or more preferred sessile *O. sulcatus* eggs compared more mobile *C. cydoniae* nymphs. Finally, sufficient alternative food resources, such as Collembola, were available at the soil level of plants. Several species of smaller staphylinids and carabids are known to feed upon Collembola (Bauer 1982, 1985, Bauer and Pfeiffer 1991, Bilde et al. 2000). The greater abundance of
alternative prey at the soil level may have retained predators and reduced the likelihood of them foraging at the foliar level. The effect of alternative prey has been documented in a number of other arthropod systems (Hazzard and Ferro 1991, Mair and Port 2001, Musser and Shelton 2003) where consumption of the focal prey by generalist predators decreased in the presence of other arthropods or other even materials, such as fungus (Dennis et al. 1991).

Several genera of ground beetles were collected from my containers; among the most abundant were Harpalus, Bembidion, and Amara. Future research should include a detailed examination of feeding preferences and foraging range for the assemblage of predators in this system. Previous research has shown that Harpalus affinis (Schr.) is primarily epigeic (Hagley et al. 1982), while research by Brust (1994) observed Harpalus pennsylvanicus in the canopy of potatoes. Bembidion is a smaller beetle and forages primarily on the ground (Koss et al. 2004). Its small size, however, may limit its ability to attack larger prey items (Frank 2004). Finally, Amara is a voracious, polyphagous ground dwelling predator (Frank 2004). It is important to identify and conserve key predators and their habits to effectively manage pests.

Previous studies showed that the addition of mulch effected beneficial arthropod and herbivore dynamics similar to my study. Reichert and Bishop (1990) and Halaj et al. (2000) increased spider abundance with a straw mulch groundcover while Brust (1994) and Johnson et al. 2004 enhanced carabid and staphylinid populations. Similar to my research, these studies found variable effects of mulch on herbivore abundance and plant damage.
Benefits of pot-in-pot planting method stem from economical analyses. Plant health and vigor is enhanced with pot-in-pot because the root zone is insulated from extreme temperature variations, and moisture content is kept under control. Plants can be harvested year long, and this method lowers labor costs associated with staking plants to prevent container blow over and the potential loss of fertilizer from container plants. The cost comparison of harvesting in the pot-in-pot method to in-field and above-ground production methods showed that the lowest cost to harvest per plant is in the pot-in-pot production method (Adrian 1998). However, the initial cost of the pot-in-pot planting method is the primary deterrent for production nursery growers (Zinati 2005). These costs include the instillation of socket pots and providing good drainage in poorly draining soil. Other negative aspects of pot-in-pot include: the potential for roots to grow into the socket pot and surrounding soil and making harvesting difficult and limited flexibility in spacing plants (Zinati 2005).

In conclusion, adding wood mulch appears to enhance the abundance of predators. The increase in alternative prey may be associated with the increase in natural enemy population. The removal of physical barriers, such as plastic pots, appears to improve the accessibility of natural enemies to the plants. Lastly, it was the soil-dwelling herbivores rather than foliar that were more susceptible to these habitat manipulations. The implications of this research to plant production nurseries are far-reaching. Habitat manipulations show a strong potential to be a successful pest management practice. However, the degree of success may vary with the plant and herbivore system (e.g. ground vs. foliar pest).
Table 1. Mean ± SEM abundance of the most common arthropod groups sampled from the ground within wood mulch or weed cloth groundcover habitats summed over four dates in 2002.

<table>
<thead>
<tr>
<th>Trophic Group</th>
<th># of Arthropods&lt;sup&gt;a&lt;/sup&gt;</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Wood Mulch</td>
</tr>
<tr>
<td>Collembola</td>
<td>1996.8 ± 394.4a</td>
</tr>
<tr>
<td>Araneae</td>
<td>63.2 ± 8.6a</td>
</tr>
<tr>
<td>Lycosidae</td>
<td>17.0 ± 2.8a</td>
</tr>
<tr>
<td>Staphylinidae</td>
<td>26.2 ± 4.4a</td>
</tr>
<tr>
<td>Carabidae</td>
<td>19.4 ± 4.3a</td>
</tr>
<tr>
<td>Formicidae</td>
<td>343.8 ± 89.1a</td>
</tr>
<tr>
<td>Gryllidae</td>
<td>180.2 ± 34.0a</td>
</tr>
<tr>
<td>Scelionidae</td>
<td>358.2 ± 110.4a</td>
</tr>
</tbody>
</table>

<sup>a</sup>Means followed by the same letter within a row are not significantly different ($P = 0.05$)
Table 2. ANOVA statistics for the abundance of arthropods on the ground within wood mulch or weedcloth groundcover habitats summed over four dates in 2002.

<table>
<thead>
<tr>
<th>Trophic Group</th>
<th>Effect</th>
<th>$F$</th>
<th>ndf, ddf</th>
<th>$P$</th>
</tr>
</thead>
<tbody>
<tr>
<td>Collembola</td>
<td>Groundcover</td>
<td>21.07</td>
<td>1, 4</td>
<td>0.01</td>
</tr>
<tr>
<td>Araneae</td>
<td>Groundcover</td>
<td>0.61</td>
<td>1, 8</td>
<td>0.46</td>
</tr>
<tr>
<td>Lycosidae</td>
<td>Groundcover</td>
<td>4.65</td>
<td>1, 4</td>
<td>0.10</td>
</tr>
<tr>
<td>Staphylinidae</td>
<td>Groundcover</td>
<td>11.18</td>
<td>1, 4</td>
<td>0.03</td>
</tr>
<tr>
<td>Carabidae</td>
<td>Groundcover</td>
<td>8.71</td>
<td>1, 8</td>
<td>0.02</td>
</tr>
<tr>
<td>Formicidae</td>
<td>Groundcover</td>
<td>0.90</td>
<td>1, 4</td>
<td>0.40</td>
</tr>
<tr>
<td>Gryllidae</td>
<td>Groundcover</td>
<td>6.02</td>
<td>1, 4</td>
<td>0.07</td>
</tr>
<tr>
<td>Scelionidae</td>
<td>Groundcover</td>
<td>1.10</td>
<td>1, 4</td>
<td>0.35</td>
</tr>
</tbody>
</table>
Table 3. Mean ± SEM abundance of arthropod groups sampled from the soil surface within containers of cotoneaster in weed cloth or mulch ground cover plots and either pot-above-ground or pot-in-pot summed over 2 dates (28 August and 13 September 2002).

<table>
<thead>
<tr>
<th>Trophic Group&lt;sup&gt;a&lt;/sup&gt;</th>
<th>Wood Mulch&lt;sup&gt;b&lt;/sup&gt;</th>
<th>Weed Cloth</th>
<th>Above Ground</th>
<th>Pot-in-Pot</th>
<th>Above Ground</th>
<th>Pot-in-Pot</th>
<th>Above Ground&lt;sup&gt;c&lt;/sup&gt;</th>
<th>Pot-in-pot</th>
</tr>
</thead>
<tbody>
<tr>
<td>Collembola</td>
<td>43.4 ± 32.4&lt;sup&gt;b&lt;/sup&gt;</td>
<td>223.0 ± 91.1&lt;sup&gt;a&lt;/sup&gt;</td>
<td>40.0 ± 23.0&lt;sup&gt;ab&lt;/sup&gt;</td>
<td>40.6 ± 11.4&lt;sup&gt;ab&lt;/sup&gt;</td>
<td>41.7 ± 18.8</td>
<td>131.8 ± 52.9</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Araneae</td>
<td>1.4 ± 0.5&lt;sup&gt;a&lt;/sup&gt;</td>
<td>4.4 ± 1.2&lt;sup&gt;b&lt;/sup&gt;</td>
<td>1.8 ± 1.6&lt;sup&gt;a&lt;/sup&gt;</td>
<td>2.4 ± 0.7&lt;sup&gt;a&lt;/sup&gt;</td>
<td>1.6 ± 0.4</td>
<td>3.4 ± 0.7</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Staphylinidae</td>
<td>0.2 ± 0.2</td>
<td>1.8 ± 0.7</td>
<td>0.4 ± 0.2</td>
<td>1.0 ± 0.4</td>
<td>0.3 ± 0.2&lt;sup&gt;a&lt;/sup&gt;</td>
<td>1.4 ± 0.4&lt;sup&gt;b&lt;/sup&gt;</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Carabidae</td>
<td>0.2 ± 0.2</td>
<td>4.0 ± 1.0</td>
<td>0.2 ± 0.2</td>
<td>1.6 ± 0.7</td>
<td>0.2 ± 0.1&lt;sup&gt;a&lt;/sup&gt;</td>
<td>2.8 ± 2.3&lt;sup&gt;b&lt;/sup&gt;</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Formicidae</td>
<td>5.6 ± 3.0</td>
<td>40.4 ± 12.7</td>
<td>5.0 ± 1.6</td>
<td>12.8 ± 4.4</td>
<td>5.3 ± 1.6&lt;sup&gt;a&lt;/sup&gt;</td>
<td>26.6 ± 7.8&lt;sup&gt;b&lt;/sup&gt;</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Gryllidae</td>
<td>0.2 ± 0.2</td>
<td>12.6 ± 3.3</td>
<td>0.2 ± 0.2</td>
<td>18.4 ± 5.9</td>
<td>0.2 ± 0.1&lt;sup&gt;a&lt;/sup&gt;</td>
<td>15.5 ± 3.3&lt;sup&gt;b&lt;/sup&gt;</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Scelionidae</td>
<td>2.2 ± 0.9&lt;sup&gt;bc&lt;/sup&gt;</td>
<td>66.4 ± 32.2&lt;sup&gt;a&lt;/sup&gt;</td>
<td>2.2 ± 1.1&lt;sup&gt;c&lt;/sup&gt;</td>
<td>13.6 ± 7.0&lt;sup&gt;b&lt;/sup&gt;</td>
<td>2.0 ± 0.6</td>
<td>40.0 ± 17.8</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

<sup>a</sup>There was no significant main effect of groundcover on any arthropod group.

<sup>b</sup>Means followed by lower case letters indicate a significant groundcover by pot position interaction.

<sup>c</sup>Means followed by the same letter within a row are not significantly different (P = 0.05).
Table 4. ANOVA statistics for the abundance of arthropods, within containers in weed cloth or mulch groundcover and either pot-above-ground or pot-in-pot containers for Summer 2002.

<table>
<thead>
<tr>
<th>Trophic Group</th>
<th>Effect</th>
<th>$F$</th>
<th>ndf, ddf</th>
<th>$P$</th>
</tr>
</thead>
<tbody>
<tr>
<td>Collembola</td>
<td>Groundcover</td>
<td>0.63</td>
<td>1, 8</td>
<td>0.4498</td>
</tr>
<tr>
<td></td>
<td>Pot position</td>
<td>5.61</td>
<td>1, 8</td>
<td>0.0453</td>
</tr>
<tr>
<td></td>
<td>Groundcover × pot position</td>
<td>6.22</td>
<td>1, 8</td>
<td>0.0373</td>
</tr>
<tr>
<td>Araneae</td>
<td>Groundcover</td>
<td>2.15</td>
<td>1, 12</td>
<td>0.1687</td>
</tr>
<tr>
<td></td>
<td>Pot position</td>
<td>10.86</td>
<td>1, 12</td>
<td>0.0064</td>
</tr>
<tr>
<td></td>
<td>Groundcover × pot position</td>
<td>4.83</td>
<td>1, 12</td>
<td>0.0484</td>
</tr>
<tr>
<td>Staphylinidae</td>
<td>Groundcover</td>
<td>0.30</td>
<td>1, 12</td>
<td>0.5921</td>
</tr>
<tr>
<td></td>
<td>Pot position</td>
<td>10.18</td>
<td>1, 12</td>
<td>0.0078</td>
</tr>
<tr>
<td></td>
<td>Groundcover × pot position</td>
<td>1.91</td>
<td>1, 12</td>
<td>0.1918</td>
</tr>
<tr>
<td>Carabidae</td>
<td>Groundcover</td>
<td>1.30</td>
<td>1, 8</td>
<td>0.2880</td>
</tr>
<tr>
<td></td>
<td>Pot position</td>
<td>17.40</td>
<td>1, 8</td>
<td>0.0031</td>
</tr>
<tr>
<td></td>
<td>Groundcover × pot position</td>
<td>2.01</td>
<td>1, 8</td>
<td>0.1945</td>
</tr>
<tr>
<td>Formicidae</td>
<td>Groundcover</td>
<td>1.52</td>
<td>1, 4</td>
<td>0.2847</td>
</tr>
<tr>
<td></td>
<td>Pot position</td>
<td>17.99</td>
<td>1, 8</td>
<td>0.0028</td>
</tr>
<tr>
<td></td>
<td>Groundcover × pot position</td>
<td>3.25</td>
<td>1, 8</td>
<td>0.1091</td>
</tr>
<tr>
<td>Gryllidae</td>
<td>Groundcover</td>
<td>0.21</td>
<td>1, 8</td>
<td>0.6617</td>
</tr>
<tr>
<td></td>
<td>Pot position</td>
<td>142.52</td>
<td>1, 8</td>
<td>&lt; 0.0001</td>
</tr>
<tr>
<td></td>
<td>Groundcover × pot position</td>
<td>0.32</td>
<td>1, 8</td>
<td>0.5852</td>
</tr>
<tr>
<td>Scelionidae</td>
<td>Groundcover</td>
<td>2.13</td>
<td>1, 4</td>
<td>0.2182</td>
</tr>
<tr>
<td></td>
<td>Pot position</td>
<td>34.51</td>
<td>1, 8</td>
<td>0.0004</td>
</tr>
<tr>
<td></td>
<td>Groundcover × pot position</td>
<td>5.56</td>
<td>1, 8</td>
<td>0.0461</td>
</tr>
</tbody>
</table>
A.

Fig. 1a. Layout of treatment plots with weed cloth and wood mulch groundcover, and pot-above-ground and pot-in-pot, and placement of pitfall traps used for sampling (represents one replicate).  1b. Diagram of pot-in-pot and pot-above-ground treatments.
Fig. 2. Mean proportion (SE) of *Corythuca cydoniae* (Hawthorn lace bug, HLB) nymphs remaining 2 days after plants were infested. Containerized plants were in beds with wood mulch and weed cloth groundcover. Within each groundcover treatment, containers were above ground or pot-in-pot. The study was repeated on three dates: a. 24 June 2002, b: 9 July 2002, and c: 5 August 2002.
Fig. 3. Mean (SE) number of *O. sulcatus* (black vine weevil, BVW) eggs remaining 1 day after 10 eggs were placed on a petri dish at the soil surface in containers of *Cotoneaster dammeri*. Containerized plants were in beds with wood mulch and weed cloth groundcover; within each groundcover treatment, containers were above ground or pot-in-pot. The study was repeated on three dates: 12, 13, 14 August 2002. There was no significant effect of date, therefore, the figure represents the pooled values for the three dates.
Chapter 2

Habitat Manipulation: Influence of Management Practices on Spatial and Temporal Predator-Herbivore Dynamics

INTRODUCTION

Managed ecosystems are often unfavorable for natural enemies because they undergo frequent disturbances (Van Driesche and Bellows 1996, Landis et al. 2000). For example, the process of urbanization or the production of agricultural crops alters the vegetational diversity and structural complexity of a habitat reducing the availability of resources necessary for natural enemy survival and reproduction (Winchester 1997, Rypstra et al. 1999, Landis et al. 2000). Other plant or crop management practices such as the application of insecticides (Raupp et al. 1992, Landis et al., Lee et al. 2001) or field tillage (Rypstra et al. 1999, Halaj et al. 2000) also disturb arthropod community composition resulting in decreased natural enemy populations. These disturbances often result in frequent herbivore outbreaks (Roland and Taylor 1997, Landis et al. 2000). Therefore, there is a need to identify management practices that are least disruptive to herbivore – natural enemy dynamics and reduce the likelihood of pest outbreaks in managed ecosystems. To do this, a better understanding of the influence of plant or crop management practices on herbivore – natural enemy community dynamics is necessary.

Two ways that management practices are known to disturb herbivore-natural enemy communities are: 1) creating habitats that are unfavorable to natural enemies (Gurr et al. 2000, Landis et al. 2000), and 2) creating physical barriers that result in herbivores being less accessible to natural enemies (Dennis and Wratten 1991, Clark et al. 1994). Habitats that favor natural enemy populations are those that provide alternative
food resources (e.g. floral resources), create refuges for survival and reproduction, and are structurally more complex (Landis et al. 2000, Shrewsbury in press). Clark et al. (1994) found that placing strips of galvanized steel inhibited the movement of carabids and spiders. As a result, herbivore feeding damage was more severe in treatments where these predators are excluded. Other studies suggest the presence of green fences or farm roads along agricultural field edges hinder the movement of natural enemies, hence the accessibility of prey in the agricultural crop.

In previous research groundcover (weed cloth and hard wood mulch) and container position (pots placed above the ground and pots sunken into the ground [pot-in-pot]) were manipulated in nursery beds. Generally, wood mulch enhanced the abundance of alternative prey and natural enemies (Carabidae and Staphylinidae) at the ground level but not at the soil level within the containers or on plant foliage. The pot-in-pot method strongly increased abundance of both groups at the ground level and the soil level within the containers, whereas pots above ground had a greater abundance of predators on the foliage. Habitat manipulations differentially affected mortality of soil versus foliar herbivores. However, herbivores on the soil in containers suffered greater mortality than those on the foliage, regardless of groundcover or pot position.

Another management practice known to effect herbivore and natural enemy dynamics is the application of pesticides. Studies have shown a decline in natural enemy abundance following treatment of plants with insecticides (Raupp 1992, Lee et al. 2001, Maloney 2003). Pesticide applications may directly or indirectly affect natural enemies. Pesticides may be toxic to natural enemies and kill them directly or pesticides may reduce...
herbivore abundance which indirectly results in a decline in natural enemy abundance (Tallamy 2004).

The above studies suggest management practices affect natural enemies and ultimately herbivore predation. To better understand relationships between management practices, herbivores and natural enemies, this study examined spatial (foliar versus soil dwelling) and temporal (day versus night) differences in predation upon herbivores and arthropod abundance and diversity on ornamental plants in a container production nursery under varying management practices. I examined three management practices commonly implemented in ornamental plant production nurseries and compared them to alternative practices for their effect on arthropod community dynamics. Management practices examined included: 1) groundcover type: black weed cloth (conventional method) and shredded hard wood mulch (alternative method); 2) pot position method: above ground (conventional) and pot-in-pot (alternative); and 3) insecticide use: acephate application (conventional) and no insecticide application (alternative).

I predict alternative management practices should provide a more favorable environment (e.g. alternative prey, refuge from predators, variable microclimates) and reduce barriers (hot, black weed cloth and plastic plant containers) to natural enemies making herbivores more accessible. Therefore, overall natural enemy abundance and herbivore predation should be greater in plots and on plants managed under alternative practices.

In addition, predator species can vary in their foraging behaviors both spatially (ground versus foliar foraging) and temporally (day versus night) (Maloney 2003). Therefore, management practices may differentially affect dynamics between predators.
and herbivores, particularly among predators that differ in their spatial and temporal
foraging habits, and herbivores that vary spatially in their life history habits (foliar versus
ground active). To examine these dynamics, predation of herbivores and natural enemy
abundance and taxa will be monitored on the foliage and on the soil surface of container
plants, both during the day and the night. I predict natural enemy abundance and
herbivore predation will be greatest on the soil surface of containers than the foliage, and
that this effect will be even greater at night than the day.

The overall objective of this study was to determine the influence of management
practices on spatial and temporal predator – herbivore dynamics. The specific objectives
were to: 1) determine the effect of management practices on the abundance of natural
enemies and alternative prey at three locations (ground, container, and foliage); 2) determine the effect of herbivore location (foliage versus soil) on predation; and 3) determine the effect of time (day versus night) on natural enemy abundance and activity.

METHODS

Study site and experimental design.

To evaluate the effect of habitat manipulations and plant management practices in
ornamental nursery systems on beneficial arthropod taxa and abundance and herbivore
predation, field studies were conducted from July 2003 to October 2004 at Marshy Point
Nursery in Chase, Maryland. The experiment was a 2 x 2 factorial split-plot design with
12 replicates. Within each replicate, there were two treatment levels of groundcover
(whole plot): hard wood mulch and weed cloth. Within each groundcover treatment,
there were two treatment levels of planting method (sub-plot): pot-above-ground and
pot-in-pot. Each replicate had two study or treatment whole plots. Treatment whole
plots were set up to simulate ornamental container plant production beds. Each treatment whole plot was 1.5 meters by 4.6 meters in size (Fig. 1). All replicates were separated by a 1.5m weed cloth buffer. Azaleas (Rhododendron spp.) were selected as study plants because of their high economic importance to nurseries and landscapes, as well as their susceptibility to pest arthropods. Several cultivars of azaleas were used in this study. These included “Delaware Valley white”, “Bixby (Weston)”, “Pink Gardenia”, and “Midnight Flare”. Cultivars were randomly assigned to study plots. Azaleas were potted in 11.7 liter containers with a sterile potting medium (Pro-Mix, Scotts-Sierra Horticultural Products Company, Ohio). In each treatment whole plot, containers were placed in five rows with three containers per row (15 plants per whole plot). Containers were separated within and between rows by 0.33 meter. Plants were fertilized in May 2003 and 2004 with Nutricote© 14-14-14 (Vicksburg Tri Pro) slow release fertilizer and watered throughout the study as needed with drip irrigation. All plots and containers were hand weeded, and the grassy areas adjacent to the plots were mowed periodically to maintain a low turf cover.

Within each replicate, one of two groundcover treatments (wood mulch or weed cloth) was randomly assigned to each treatment whole plot. The mulch was shredded hardwood mulch (mean shredded length 7.6 cm) and was spread across the plot at a depth of 5 cm deep on top of a black weed cloth groundcover (Polypropylene, SI Performance Technology, Ringgold, GA). The other plot was covered with black weed cloth only. Pot position sub-plot treatments (pot-in-pot and pot-above-ground) were randomly assigned within each groundcover whole plot treatment. Six plants in half of the plot were assigned to pot-in-pot; the remaining nine plants were assigned to the pot-above-
ground treatment. The pot-in-pot planting method is a relatively new technique used by ornamental nursery growers. With the pot-in-pot method, an empty container called the socket pot is sunk in the soil with the pot rim at soil surface level. A containerized plant is then placed in the submerged socket pot. In the above ground method, more commonly used by plant producers, containerized plants are lined out in rows and set on the surface of the groundcover.

**Arthropod taxa and abundance.**

**Influence of Groundcover and Pot Position.**

*Activity at the ground level.* Sampling was conducted by placing four pit fall traps in each of the study whole plots. Two traps were placed in the pot-in-pot sub-plot; the other two were placed in the pot-above-ground sub plot (Fig. 1). Pit fall traps were as described in Chapter 1. In 2003, pitfall sampling was conducted once 6 August. In 2004, sampling was conducted three times: 18 May, 16 July, and 10 September. Traps remained in the plots for 7 to 10 day intervals. To standardize arthropod counts, data is presented as number of arthropods per day. Traps collected in the same sub-plot treatment were pooled, returned to the laboratory, rinsed, and stored in 70% ethyl alcohol. At a later date arthropods were sorted and identified to family level.

*Activity within plant containers.* Sampling was conducted using pitfall traps that were set up inside the containers as described in Chapter 1. The same sampling dates and duration occurred for the container traps as the ground traps. Collection, storage, and identification of samples were the same as above.

*Activity at the foliar level.* Sampling was conducted using timed visual counts and beat-sampling on the azalea foliage (2004 only). Timed visual counts were
conducted during the day on azaleas that had been infested previously with azalea lace bug, *Stephanitis pyrioides* Scott (see following section). Two infested plants were selected at random in each groundcover - pot position treatment combination. Three timed visual counts of arthropods on the foliage were conducted: 9 June, 16 July, and 16 September 2004. The first two counts were done with three minutes per plant while the last count was done with five minutes per plant. Data collected on observed arthropods included abundance, and identification to family and trophic group. The data are presented as the number of arthropods per minute.

Beat-sampling of azalea foliage was also conducted but during the day and night to determine the effect of groundcover and pot position on arthropod activity and if arthropod activity differed temporally. Three plants were selected at random from both pot-in-pot and pot-above-ground containers in both groundcover treatments. Each plant was struck with a rod 20 times (× 3 plants = 60 beats per treatment combination) over a tray that contained a film of 70% ethyl alcohol. Any arthropods that fell in the tray were washed in to storage jars for later identification. Two beat-samplings occurred in 2004: a day sampling on 23 September and an evening sampling on 1 October.

**Influence of insecticide on arthropods.**

Additional plots designed and maintained in an identical manner to the weed cloth as described above were sprayed twice with the insecticide Orthene® (acephate) on 8 May and 16 July 2004. This treatment determined the effect of insecticides on natural enemies at the ground, container, and foliar level on a weed cloth surface. At the foliar level, the effect of insecticide and pot position were also examined. Pitfall sampling of the ground and container active arthropods and beat-sampling of the foliar active arthropods, and
collection, storage, and identification of samples was conducted as described above. Beat-sampling was conducted during the day and night to determine if insecticides varied in their effect on arthropods that were active in the day versus the night.

**Herbivore Survival.**

*Herbivore survival trials 2003.* Beet armyworm, *Spodoptera exigua* Hubner, larvae were used as a model herbivore to study the effect of ground cover and pot position on the survival of herbivores active in the plant canopy compared to the soil. *Spodoptera exigua* was selected for its ease to manipulate and availability through a commercial supplier. In each groundcover – pot position treatment plot, *S. exigua* larvae were placed on one randomly selected plant was. Second and third instar larvae were pinned with size 0 insect pins through their last abdominal segment. Preliminary trials found *S. exigua* could survive a minimum of 48 hours pinned condition (S. Frank, Pers. comm.). One larva was pinned to the soil, and another larva was pinned to the foliage of each study plant. This experiment was repeated three times in 2003: (I) 30-31 July, (II) 6-7 August, and (III) 7-8 August. Larvae in trial I were recorded as 1) present and alive or 2) missing or dead 24 hours after pinning on plants. Larvae in trial II were monitored five hours after pinning. In trial III, larvae were monitored four hours after pinning. Differences in sampling intervals between trials was due to increasing rates in *S. exigua* mortality in later trials. All trials were monitored until there was approximately 50-65% mortality or disappearance of the *S. exigua* cohorts.

*Herbivore survival trials 2004.* Two plants from each plot within each treatment combination were randomly infested with *S. pyrioides* at the start of the 2004 growing season on 8 May. *Stephanitis pyrioides* were field collected from a single population in
Glenn Dale, MD. To infest plants, *S. pyrioides* were released from two open vials, each with 25 5th instar nymphs, placed at the base of the study plants to allow nymphs to freely move onto plants. Three post counts were conducted over the season on *S. pyrioides* abundance. Post counts took place on 10 May (day two after infestation), 26 May (day 18 after infestation), and 16 September. Post counts consisted of a five-minute visual search for *S. pyrioides* on each infested study plant.

Studies were conducted in 2004 using black cutworm, *Agrotis ipsilon* Hufnagel, larvae. *Agrotis ipsilon* were used because they were easy to manipulate in a field setting and available from a supplier. *Agrotis ipsilon* eggs were donated by Dow AgroSciences (Indianapolis, Indiana) and maintained in the laboratory in a growth chamber (20ºC, 12:12 D:L) on artificial diet. Second and third instar larvae were pinned through the last abdominal segment. One plant was selected in each pot position within each ground cover treatment. Each plant had five larvae pinned to random locations on the soil surface of the container and five larvae pinned randomly to the foliage of the plant. Two trials were conducted: (I) 3-4 September and (II) 4-5 September. Trial I was during the day; *A. ipsilon* larvae were pinned to plants at 9 a.m. and sampling of *A. ipsilon* survival took place every two hours for eight hours and again the next morning. Trial II was conducted at night; larvae were pinned to the plants at 8 p.m and sampling of *A. ipsilon* survival occurred every two hours for eight hours and again the next morning. Approximately 65% mortality of all *A. ipsilon* cohorts occurred at the hour 4 sampling. Mortality after 4 hours was high and treatment differences were less likely to be detected. Therefore, hour 4 survival data are presented.
**Spatial and Temporal Dynamics of Predators.**

To further examine spatial and temporal differences in predators and their activity, visual sampling was conducted during the day and night of *A. ipsilon* larvae on the foliage and on the soil. The effect of groundcover and pot position on spatial (soil versus foliage) and temporal (day versus night) differences in the frequency of predation and the taxa of predators attacking *A. ipsilon* was recorded. Visual observations of predation were made concurrently with sampling for survival of *A. ipsilon* larvae every described above. Predation event data was summed over the duration of each day and night study (= 5 observations for each study). The frequency of observed predation events was totaled for each replicate, groundcover, pot position, and herbivore location. Predators feeding on *A. ipsilon* were identified to family to determine key predators in this study system and to calculate and compare predator family richness between day and night trials.

**Statistical Analysis.** The abundance and diversity of arthropod taxa on the ground, within the containers, and on the foliage was analyzed using an analysis of variance (ANOVA) for a split plot factorial design (Proc Mixed, SAS Institute 1999). Herbivore survival data was also analyzed in the same manner (Proc Mixed, SAS Institute 1999). Arthropod data were pooled across sampling dates. For variables where temporal (day versus night) effect were examined, a repeated measures analysis (Proc Mixed, SAS Institute 1999) was used. To determine if data met the assumptions of ANOVA, homogeneity of variances and normality were examined using Proc Univariate and plotting residuals (SAS Institute 1999). Data that did not meet the assumptions of normality and homogeneous variances were log_{10} transformed or variances were
partitioned and analysis was performed on transformed or partitioned data. Arthropod
data that was not successfully transformed was then analyzed using the Shierer-Ray-Hare
non-parametric method, a rank test (Sokal and Rohlf 1995). Data are presented as
untransformed means ± SEM. Differences were considered significant at $P < 0.05$. The
survival data for *S. exigua* was analyzed using Proc Freq (SAS Institute 1999) with the
chi-square and Fisher’s exact test options. The comparisons of particular interest were
between groundcover types, pot positions, and herbivore locations. The differences in
observed predation counts were analyzed using Proc Mixed (SAS Institute 1999). For the
more abundant families of predators (ants and spiders), Proc Means (SAS Institute 1999)
was used to calculate the average abundance for each groundcover, pot position and
herbivore location. The frequency of predation was observed at five times during the day
and five times at night. In order to determine differences in family richness, the
treatments in this analysis were either day or night, and the average number of families
counted per replicate in each treatment was calculated in Proc Means (SAS Institute
1999). Data did not meet the assumptions for ANOVA analysis. Therefore, statistical
differences in the data were determined using a non-parametric analysis (Proc Npar1way,
SAS Institute 1999).

**RESULTS**

**Arthropod Taxa and Abundance.**

**Influence of Groundcover and Pot Position.**

*Activity at the ground level.* In 2003, there were no significant main effects or
interactive effects of groundcover on Collembola (Tables 1 and 2). Of the four most
abundant natural enemy taxa, there was a significant effect of groundcover on
Staphylinidae and Carabidae abundance (Tables 1 and 2), where there was a higher abundance of both of these groups in wood mulch (Staphylinidae: 0.6 ± 0.1 Carabidae: 0.4 ± 0.1) than weed cloth (Staphylinidae: 0.3 ± 0.07 and Carabidae: 0.1 ± 0.05). There was a significant groundcover by pot position interaction ($F = 6.2$, df = 1, 39, $P = 0.02$) on Araneae abundance; more spiders were captured in the pot-in-pot treatment sub-plots, especially in the wood mulch plots (Tables 1 and 2). There was no significant main or interactive groundcover or pot position effects on any of the other arthropod groups examined in 2003 (Tables 1 and 2).

Collembola, a potential alternative prey item, had no main or interactive effect of groundcover or pot position on Collembola abundance (Tables 3 and 4). Within the natural enemy group, Araneae abundance was significantly influenced by pot position; more spiders were captured on the ground in the pot-in-pot sub-plot (1.4 ± 0.2) of the plot than above ground (0.9 ± 0.1) (Tables 3 and 4). There was a significant interactive effect of groundcover and pot position on Carabidae and Formicidae with both insect families having a higher abundance on the ground in the pot-above-ground areas of the plot especially in wood mulch (Tables 3 and 4). Ground cover and pot position had a marginally significant interactive effect ($P = 0.08$) on Staphylinidae abundance.

Activity within plant containers. The effect of ground cover and pot position on arthropod taxa and abundance at the soil level within plant containers, results were similar in 2003 and 2004. The most abundant taxa of alternative prey were Collembola and natural enemies were Araneae, Staphylinidae, Carabidae, and Formicidae (Tables 5 and 7). There was no significant interaction between groundcover and pot position, and no significant main effect of groundcover on abundance of any arthropod taxa for both
years, though abundance was generally greater in the presence of mulch (Tables 5, 6, 7, and 8). However, for all arthropod taxa examined, abundance was consistently greatest in containers in the pot-in-pot method, regardless of groundcover type (Tables 5, 6, 7, and 8). The only exception to this pattern was in 2004 where there was a marginally significant ($P = 0.08$) interaction between groundcover and pot position on Carabidae abundance with more beetles captured in the pot-in-pot containers, but more so in the wood mulch compared to weed cloth plots (Tables 7 and 8).

*Activity at the foliar level.* The most abundant family found while visually searching azalea foliage was Araneae. There were no significant main effects or interactive effects of groundcover or pot position on spider abundance. Though there were numerically more spiders on pot-above-ground containers (2.4 ± 0.2) than pot-in-pot (2.1 ± 0.2).

As in the visual sampling of foliage, spiders were the most abundant natural enemy family during the day and night sampled while beating azalea foliage. There was no time effect on spider abundance ($F = 2.0$, df = 7, 51.1, $P = 0.09$), and only pot position significantly affected spider abundance during both time periods (Figs. 2a and 3a) with a greater abundance on foliage from plants in pot-above-ground containers (Day: 24.0 ± 1.6; Night: 24.4 ± 1.4) than pot-in-pot (Day: 18.9 ± 1.8; Night 18.5 ± 1.9). Groundcover did not significantly alter the abundance of the spiders or any of the spider families during the day. The abundance of aranaeids was significantly affected by pot position (Fig. 2c). There was a significant interaction between groundcover and pot position for salticid spiders (Fig. 2b). These spiders were also more abundant in pot-in-pot containers but especially in the weed cloth plots. At night, there was no effect of groundcover on
spider abundance except for Thomisidae where there were more crab spiders in wood mulch plots than weed cloth (Fig. 3c). Also, there was no significant effect of pot position except for spiders as a group with more spiders sampled in pot-in-pot containers (Fig. 3a).

Influence of insecticide on arthropods.

Natural Enemies

*Activity at the ground level.* Insecticide applications did not significantly affect the abundance of arthropod families compared to plots that were unsprayed (Tables 9 and 10).

*Activity within containers.* Abundance of one of the more common natural enemy families were affected by insecticide applications when comparing treated and untreated plants: Carabidae. Carabidae had a higher abundance on plants in insecticide treated plots (0.4 ± 0.1) than untreated plots (Carabidae: 0.2 ± 0.04) (Tables 11 and 12).

*Activity at the foliar level.* Using a repeated measures analysis, there was a significant interactive effect of date and all treatment combinations ($P = 0.01$). There were fewer spiders on insecticide treated (15.8 ± 2.0) than untreated (21.8 ± 1.9) on plants during the day, however this effect was not significant ($P = 0.09$). Pot position did not effect spider abundance during the day ($P = 0.34$). At night, there were significantly fewer spiders on treated plants (12.3 ± 1.7) than untreated (23.2 ± 1.9) ($F = 12.4$, df = 1, 27, $P = 0.002$). Also there were fewer spiders on the foliage of pot-in-pot (17.2 ± 2.7) containers than pot-above-ground containers (21.8 ± 1.8) ($F = 6.1$, df = 1, 27, $P = 0.02$).
Herbivores

*Stephanitis pyrioides* abundance at the end of the season in September did not differ between insecticide and non-insecticide treated plots ($F = 1.1$, df = 1, 26.2, $P = 0.31$). However, abundance of *S. pyrioides* was numerically lower on insecticide treated plants ($12.8 \pm 4.7$) than plants that were untreated ($21.5 \pm 7.5$).

**Herbivore survival.**

*Herbivore survival trials 2003.* The main effects of groundcover, pot position, and herbivore location had varying effects on *S. exigua* mortality in the three predation trials (Table 13). There was no significant effect on mortality when comparing pot-in-pot and pot-above-ground in any of the three trials (Trial 1: $P = 0.21$, Trial 2: $P = 0.28$, Trial 3: $P = 1.0$). In trial 1, there was a significant difference between wood mulch and weed cloth groundcovers (Table 13) where more herbivores placed on plants in the wood mulch plots died (36 dead) compared to larvae in the weed cloth plots (24 dead). In trials 1 and 3, there was a significant effect of herbivore location, soil versus foliage, on mortality (Table 13). Mortality of herbivores pinned to the soil was higher compared to larvae on the foliage (Trial 1: 39 dead on soil, 21 dead on foliage, Trial 3: 35 dead on soil, 19 dead on the foliage).

*Herbivore survival trials 2004.*

*Stephanitis pyrioides.* Abundance of *S. pyrioides* was monitored on three dates after initial infestation on 8 May: 10 May, 26 May, and 16 September 2004. A repeated measures analysis showed that there was no significant main effect of habitat manipulations on *S. pyrioides* abundance over the growing season ($F = 0.9$, df = 3, 44, $P$
= 0.47). However, there was a significant interaction of date and treatment ($F = 3.1$, df = 6, 74.7, $P = 0.01$) and a main effect of date ($F = 25.9$, df = 2, 65.2, $P < 0.01$).

There were no significant main effects of groundcover and pot position ($F = 1.0$, df = 1, 39, $P = 0.32$ and $P = 1.7$, df = 1, 39, $F = 0.20$, respectively) or an interaction of either effect ($F = 0.2$, df = 1, 39, $P = 0.66$) on $S. pyrioides$ abundance on 10 May (Fig. 4). On 26 May, there was a marginally significant effect of groundcover ($F = 4.5$, df = 1, 5, $P = 0.09$) and a significant effect of pot position ($F = 6.9$, df = 1, 34, $P = 0.01$) and no interaction ($F = 0.03$, df = 1, 34, $P = 0.86$) (Fig. 4). Abundance of $S. pyrioides$ was lower on plants in wood mulch habitats (6.2 ± 0.9) than weed cloth (12.2 ± 2.2). There was a significantly lower abundance of $S. pyrioides$ on plants in pot-above-ground containers (7.0 ± 1.5) than pot-in-pot containers (11.4 ± 1.9) (Fig. 4). At the end of the season on 16 September, there was no significant effect of groundcover ($F = 0.5$, df = 1, 9.9, $P = 0.52$), however, there was a marginally significant effect of pot position ($F = 3.4$, df = 1, 33.1, $P = 0.07$) with fewer $S. pyrioides$ sampled on pot-above-ground containers (14.9 ± 5.1) than pot-in-pot (22.0 ± 6.1) (Fig. 4).

$Agrotis ipsilon$. There was a significant interaction of time and all treatment combinations (groundcover type, pot position method, and herbivore location) ($F = 8.6$, df = 15, 126, $P < 0.01$) for the proportion of $A. ipsilon$ mortality. However, most of this interaction is attributed to the main effect of herbivore location (soil versus foliar), where there was a significantly higher proportion of $A. ipsilon$ mortality on the soil (0.77 ± 0.03) than the foliage (0.33 ± 0.03). For both trials, there was no significant main effect of groundcover, so the results were pooled across groundcover tactics. During the daytime, $A. ipsilon$ mortality was not significantly affected by pot position or any other interactive
effects. However, mortality was significantly influenced by herbivore location \((F = 36.7, df = 1, 83, P < 0.01)\). A higher proportion of \textit{A. ipsilon} died on the soil \((0.8 \pm 0.04)\) compared to the foliage \((0.4 \pm 0.04)\) (Fig. 5a). At night, there was no significant effect of pot position on \textit{A. ipsilon} mortality (Fig. 5b). However, there was a main effect of herbivore location \((F = 85.7, df = 1, 74.2, P < 0.01)\) and a significant interaction of pot position and herbivore location \((F = 13.6, df = 1, 74.2, P < 0.01)\). More \textit{A. ipsilon} died on the soil \((0.8 \pm 0.05)\) than the foliage \((0.3 \pm 0.04)\). \textit{Agrotis ipsilon} mortality on the soil was enhanced when the containers were pot-in-pot \((0.9 \pm 0.04)\) compared to above ground \((0.6 \pm 0.07)\) (Fig. 5b).

**Spatial and Temporal Dynamics of Predators.**

There was no significant main effect of groundcover for either day or night trials on predation events (day: \(F = 0.5, df = 1, 10, P = 0.49\); night: \(F = 0.2, df = 1, 83, P = 0.70\)), so values were pooled across groundcover. During the day trial, there were not any significant main effects or interactions, however, predation events were numerically greater when herbivores were placed on the soil \((F = 3.4, df = 1, 78, P = 0.07)\) (Fig. 6a). At night, there was a significant main effect of pot position \((F = 4.7, df = 1, 83, P = 0.03)\) and herbivore location \((F = 4.7, df = 1, 83, P = 0.03)\). More predators were observed attacking \textit{A. ipsilon} in the pot-above-ground containers \((1.8 \pm 0.2)\) than pot-in-pot \((1.1 \pm 0.2)\). Predation also was greater at the soil level \((2.0 \pm 0.3)\) compared to the foliage \((0.9 \pm 0.1)\) (Fig. 6b).

Several families of predators were observed frequently during these trials. They were ants (Hymenoptera: Formicidae), jumping spiders (Araneae: Salticidae), and anyphaenid spiders (Araneae: Anyphaenidae). The frequency of predation on \textit{A. ipsilon}
by each of these predator families varied among treatments. For the day predation trial, salticid spiders were most abundant on the foliage (1.1 ± 0.1), while ants dominated on the soil (0.9 ± 0.2) (Fig. 7a). These predators were not affected by groundcover type or pot position. Ants also were a dominant predator on the soil during the night trial with an overall increase in abundance as well (1.7 ± 0.3). At night, anyphaenid spiders were the most abundant predator on the foliage (0.6 ± 0.1) (Fig. 7b), whereas salticids were nonexistent. Interestingly, groundcover or pot position did not appear to affect predation acts during the day, an opposite trend occurred at night. Anyphaenids were more abundant on the foliage of containers in wood mulch habitats (0.9 ± 0.2), regardless of pot position, compared to weed cloth (0.3 ± 0.1). Ants on the soil were influenced slightly by pot position, with more ants observed on the pot-above-ground containers (2.2 ± 0.4) than the pot-in-pot containers (1.3 ± 0.4) regardless of groundcover.

There was a significant difference in family richness between day and night *A. ipsilon* predation trials ($\chi^2 = 8.8, P = 0.003$). There was greater family diversity of predators active at night (3.33 ± 0.38) compared to the diversity of day active predators (2.08 ± 0.08) (Fig. 8).

**DISCUSSION**

In general this study provided support for my prediction that natural enemy taxa and predation upon herbivores would be greater in plots and on plants under alternative (wood mulch groundcover, pot-in-pot planting methods, and no insecticide applications) compared to conventional management practices in a production nursery, although this pattern was not consistent for all families of arthropods or herbivores examined.
This study also provided support for my prediction that predation upon herbivores would be greater on the soil of the container plants than on the foliage and that this effect would be greater at night than the day. Of interest and counter to my prediction, more natural enemies were seen on the foliage of container plants.

The effects of groundcover varied with arthropod families active at the ground level between both years. Moreover, plants in the pot-in-pot system consistently had a greater abundance of taxa at the soil surface within the containers than containers in the above ground system regardless of groundcover type for both 2003 and 2004. Only for Collembola was this effect slightly enhanced in mulched plots.

The most abundant taxon of alternative prey was Collembola. However, unlike the previous study in 2002 (Chapter 1), wood mulch did not significantly enhance collembolan population size. It is difficult to determine why Collembola abundance was higher in 2002 and not in 2003 or 2004, even with the addition of mulch. One possible explanation is that weather was unseasonably hot with low precipitation in 2002 (37.13 inches). Whereas, rainfall patterns in 2003 and 2004 were nearly the opposite (2003 = 62.66 inches; 2004 = 45.67 inches). Mulch has been shown to enhance soil-level moisture in periods of drought such as 2002, and might sustain enough moisture to have enhanced populations of Collembola. These arthropods are known to favor habitats with higher moisture (Frampton et al. 2000). Whereas, moisture was more abundant in 2003 and 2004 which may have resulted in conditions that were favorable to Collembola. Therefore, mulch may not have been as suitable to population build-up or enhance population growth for Collembola in wet years (2003 and 2004) as in a dry one (2002).
Several natural enemy families showed similar trends in 2003 and 2004 compared to previous research in 2002 (Chapter 1). Results of 2003 staphylinid and carabid abundance were identical to results in 2002 where significantly more staphylinid and carabid beetles were captured in wood mulch plots, while in 2004, the effects of groundcover type was dependent more on pot position. Since potential alternative prey taxa (Collembola) did not differ between either groundcover type for 2003 but was greater in mulched plots in 2004, it is possible that these ground dwelling natural enemies simply preferred a more complex ground habitat (wood mulch) with more hiding places from other predators than the simple weed cloth habitat (Landis et al. 2000, Shrewsbury in press). Mulch could also possibly help to moderate hot summer temperatures and enhance relative humidity at the soil level. Both of these factors have been shown to enhance natural enemy abundance (Landis et al. 2000).

Groundcover and pot position treatments had variable effects on arthropods. Araneae in 2003 was significantly more abundant in wood mulch plots, especially in sub-plots that were pot-in-pot. In 2004, there was no interaction or main effect of groundcover, however, more spiders were captured in pot-in-pot sub-plots. In 2002 mulch did not enhance populations of spiders. In fact, spiders, especially Lycosidae, had a higher abundance in weed cloth plots (Chapter 1). I suggested that these spiders might have been tracking a preferred prey item, field crickets (Orthoptera: Gryllidae) in weed cloth plots. However, there were fewer field crickets in the plots in 2003 and 2004. The enclosed denser canopy of azaleas in the pot-in-pot section might have provided a favorable microhabitat structure and additional hiding niches for ground dwelling spiders (Uetz 1991, Rypstra et al. 1999).
This study, in addition to research in Chapter 1, is one of the first that provides evidence that plastic containers act as a physical barrier to natural enemies and inhibit access to plants and the potential resources (prey) in containers. For each natural enemy taxa examined, abundance was consistently greater in containers in the pot-in-pot planting system. Plant containers in this study, and in most container production nurseries, are composed of plastic. Plastic substances are sometimes used in studies to exclude ground predators. For example, Dennis and Wratten (1991) used polyethylene (a waxy plastic) circles in cages as a deterrent for staphylinid movement.

Potential alternative prey taxa, such as Collembola, were collected most often in pot-in-pot containers regardless of groundcover in both 2003 and 2004. Other research has shown that containers in pot-in-pot production are up to 13°C cooler at the root zone level than above ground containers (Ruter 1993). In addition, pot-in-pot production promotes moisture content control with higher moisture levels than containers that are above ground (Zinati 2005). Cooler temperatures and higher moisture are characteristics that are important for Collembola survival and would enhance their abundance (Frampton et al. 2000).

Interestingly, the positive response by staphylinids and carabids to the addition of mulch at the plot level did not transfer to any significant interactive effect between mulch and pot position at the soil level within containers in 2003. Staphylinids and carabids were more abundant in containers in the pot-in-pot planting system, regardless of the groundcover type present in the plot. In 2004, there was a marginally significant interaction when carabids were captured more often in pot-in-pot containers but more so in mulch. Carabids could more easily access plant containers because there was higher
carabid capture in ground traps when containers were above ground. The plastic containers likely were prohibiting movement of carabids into the containers.

On the other hand, Araneae did differ in abundance at the plot level between plots with mulch and weed cloth in 2003, but not in 2004. In both years, spiders were more abundant in pot-in-pot containers. However, groundcover did not significantly influence the abundance of spiders within containers, though spider abundance was generally higher in the presence of wood mulch. It is likely that ground spiders in this system were not good climbers of the plastic containers. When this barrier was removed (e.g. pot-in-pot system), it appears specific spider taxa could access the plants.

The effects of groundcover and pot position treatments were similar between day and night for spiders. That is, spiders were nearly equal in abundance, despite treatments for both day and night. Spiders were more abundant in the foliage of pot-above-ground containers as determined by beat-sampling. Groundcover did not enhance the abundance of these spider families with the exception of Thomicsdae. This is perhaps the result of the fact that these spiders are foliar dwellers. Spiders may be more abundant on the foliage of pot-above-ground containers as the result of prey specialization. The groundcover type, mulch or weed cloth, may not have affected the preferred prey of these predators. Maloney et al. (2003) discusses the concept that spider prey specialization is often more common than realized. Spiders, as a whole, therefore, show a strong numerical response to prey density (Riechert and Lockley 1984) and will migrate from decreasing prey density patches to those with higher prey densities (Riechert and Lockley 1984, Harwood et al. 2001). Spider density might have varied by pot position for several reasons. Previous work has shown that various salticid species feed on diverse
groups, such as ants, flies, and lepidopteran larvae (reviewed by Maloney et al. 2003). My original prediction was that the plastic container acted as a barrier to predators and would inhibit their movement into the container and on to the plant. However, these foliar spiders may have had the ability to climb plastic, or they were transported to the higher foliage via wind.

The application of insecticides had interesting effects on arthropod abundance. I had predicted that insecticide application would adversely affect predator abundance. However, two groups of predators, Carabidae and Araneae, were captured more often from insecticide treated containers. It is possible that the application of Orthene® caused sublethal effects on insects. The irritation caused by the spray could have increased carabid and spider activity within the containers. This could increase the chance of trapping and overestimating the abundance of both groups.

Arthropod activity in the foliage as a response to insecticide application was what I predicted. Other studies have shown that insecticide use, while intended to reduce pest densities, also disrupt natural enemy complexes (reviewed by Raupp et al. 1992). The most dominant foliar predator in this system was Araneae, and its abundance was significantly less on insecticide treated plants. Many studies conclude that spiders have increased sensitivity to some pesticides, such as synthetic pyrethroids, organophosphates, and carbamate (reviewed by Maloney et al. 2003). Orthene® is a systemic chemical that should limit population growth of those arthropods that consume foliage. In this system, the target herbivore of the insecticide was also numerically less abundant on treated azaleas. If *S. pyrioides* abundance was lower than other plants in this study, spider density might be lower not because of direct mortality, but because prey density was
lower on treated plants. Spiders responded numerically to prey density (reviewed by Maloney et al. 2003).

I found variable and unexpected results of habitat manipulations on the foliar herbivore, *S. pyrioides*. Following *S. pyrioides* cohorts 18 days after their release, there was slightly lower abundance in the wood mulch than the weed cloth, but there was a lower abundance in pot-above-ground containers. With the use of repeated measures analysis, we were also able to determine that the effect of my habitat manipulations varied over time. Ultimately, at the end of the season, *S. pyrioides* abundance was lowest on containers above ground. This result if consistent with the observation that significantly more predators, especially spiders, were found in the canopy of plants that were above ground. One of the families of spiders sampled was Anyphaenidae. Shrewsbury (*in press*) showed that *S. pyrioides* abundance was lower in complex habitats where a key predator, *Anyphaena celer* (Araneae: Anyphaenidae) was common.

More *S. exigua* than expected died when placed on containers in the wood mulch plots than weed cloth for one trial. This result concurs with the prediction that the addition of mulch would enhance the abundance of natural enemies and cascade to increase mortality of herbivores. Natural enemy populations were enhanced with wood mulch in 2003 while this trial took place. However, the other two trials did not show a significant effect of groundcover. *Spodoptera exigua* mortality was also higher on the soil compared to the foliage regardless of pot position in two trials. Predators that attacked *S. exigua* were possibly arthropods that could climb the plastic barrier and then foraged within the container.
Surprisingly, there was no main effect of ground cover for either trial that examined my treatments on the mortality of *A. ipsilon*. I had predicted that attracting more ground dwelling predators, such as carabids and spiders, to wood mulch plots would have resulted in higher *A. ipsilon* mortality. However, there was a significant herbivore location or an herbivore location by pot position interaction during the day and night trials. There was higher mortality of *A. ipsilon* on the soil than foliage, and this effect was enhanced in pot-in-pot containers at night. Results from 2002 also showed that there was increased *O. sulcatus* egg mortality in pot-in-pot containers (Chapter 1). Why the mortality of *A. ipsilon* on the soil was enhanced at nightfall in pot-in-pot containers is uncertain. There was a greater diversity of night active predators, and possibly many of these families were ones that did not climb or chose not to climb plant containers, for example field crickets (Orthoptera: Gryllidae). With containers submerged into the ground, predators gained access to the plant and foraged more easily than those containers that were above ground. During the day, active predators were those that could potentially climb containers more easily, therefore plastic containers were not a serious barrier as also seen in the day trials using *S. exigua*.

The data examining the mortality of both *S. exigua* and *A. ipsilon* is important for another reason. It sheds some light on not only where predators forage, but it also reveals differences in when they forage. There were no significant trends in observed predation events during the daytime. At night, however, there were more predation events observed overall, and more so on the soil surface of containers, especially in pot-above-ground. These predation trends, however, were slightly different than what we would have predicted based upon *A. ipsilon* mortality data. More *A. ipsilon* were dead on the
soil, but in pot-in-pot containers, not pot-above-ground. However, observations of predation took place every two hours of the study and significant amounts of predation could have taken place between sampling.

Dominant predator families were Formicidae (ants), Salticidae (jumping spiders), and Anyphaenidae (a hunting spider). Very interestingly, ants and spiders divided their foraging domain within the plant. Ants were seen attacking *A. ipsilon* on the soil, while spiders dominated predation in the foliage. Salticid spiders dominated foliar predation events during the day while anyphaenids were found most often at night. Both spider families, despite occupying the same spatial niche, were able to temporally divide when they foraged. This is an important discovery as both are active hunting spiders and could potentially compete for the same herbivore resources, especially if spider densities are high (Maloney 2003). If both families foraged during the same time period, it is possible that interspecific competition or intraguild predation between families could take place (Marshall and Rypstra 1999). In having this assemblage of spiders present in the azalea foliage both day and night, there, ultimately, is a reduction in prey densities. *Stephanitis pyrioides* had significantly lower densities at the end of the season on plants in above ground containers. This directly corresponds to the higher densities of foliar spider families.

The family richness of predators observed feeding on *A. ipsilon* was significantly different between day and night. More predator families were active at night than day. Other studies have also examined predator activity at night. Johnson et al. (2004) evaluated nocturnal predators and found that assemblages were slightly different from those that were day active. During the day ladybeetles and spiders were observed most
often, while at night, spiders, carabids and crickets were seen (Johnson et al. 2004). Johnson et al. (2004) also found a greater family richness at night. It is important when implementing measures that might disrupt predator assemblages to consider those animals not only active during the day but those active at night as well.

In conclusion, wood mulch enhanced the abundance of most ground dwelling arthropod taxa. Pot position also had interactive effects with groundcover type and enhanced certain groups, such as spiders and ants in the pot-in-pot method. For arthropods foraging within containers, pot-in-pot captured significantly more alternative prey and natural enemy families than containers above ground. This trend occurred regardless of groundcover type. Natural enemy activity on the ground that had increased in the presence of wood mulch did not correspond to an increase in natural enemy abundance within containers in those plots. Groundcover did not influence the abundance of natural enemies found in the azalea foliage, but more natural enemies, especially spiders, were found on pot-above-ground containers. The effect of an insecticide application did not significantly alter the abundance of arthropod families on the ground. There were more variable results of predator abundance in containers treated with insecticide, especially for carabids and spiders. However, natural enemies, especially spiders, were significantly less on azalea foliage that had been sprayed.

Herbivore abundance and mortality also varied with the habitat manipulations. *Stephanitis pyrioides* had an overall lower abundance on plants in wood mulch plots during the midseason and fall counts, and with significantly lower abundance on plants in above ground containers. In examining spatial differences in herbivore mortality, *A. ipsilon* had greater mortality on the soil compared to the foliage regardless of
groundcover type. Temporal differences in *A. ipsilon* mortality showed that mortality on
the soil was enhanced by pot-in-pot at night. Predator diversity was also greater at night
than during the day. Predators were observed most often on the soil and containers that
were above ground. Moreover, the foliar spider assemblage changed between night and
day with two hunting spider families consuming similar herbivore resources.

Future research should examine the effects of these manipulations on different
herbivore and predator systems. From this study, herbivores that occupy different spatial
niches had variable abundance and mortality. Predators on the ground, within the
containers, and on the foliage also varied in their abundance with respect to groundcover
type and pot position. Understanding the links between enhancing alternative prey and
natural enemies and the effects on herbivore populations are critical to the successful
implementation of these tactics in production nurseries. Growers need to identify key
herbivore pests and implement tactics that will enhance the natural enemies able to
suppress that pest. For example, mortality on soil dwelling herbivores, such as *O.
sulcatus* (Chapter 1) and *A. ipsilon*, has been shown to be greater in pot-in-pot containers.
This corresponds with the greater abundance of ground dwelling predators captured
within containers. Foliar herbivores have more variable mortality with respect to pot
position. Types of mulch should be researched further in order to attract the optimal
numbers of predators. The addition of wood mulch and pot-in-pot containers has the
potential to be an effective tool to manage herbivores in ornamental production nurseries.
Table 1. Mean ± SEM abundance of the most common arthropod groups sampled from the ground within wood mulch and weed cloth groundcover habitats for 6 August 2003.

<table>
<thead>
<tr>
<th>Arthropod Group</th>
<th>Number of Arthropods&lt;sup&gt;a&lt;/sup&gt;</th>
<th>Wood Mulch&lt;sup&gt;c&lt;/sup&gt;</th>
<th>Weed Cloth</th>
<th>Wood Mulch&lt;sup&gt;d&lt;/sup&gt;</th>
<th>Weed Cloth</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>Above Ground</td>
<td>Pot-in-pot</td>
<td>Above Ground</td>
<td>Pot-in-pot</td>
</tr>
<tr>
<td>Collembola</td>
<td>24.7 ± 7.1</td>
<td>25.1 ± 4.7</td>
<td>24.3 ± 3.4</td>
<td>22.5 ± 4.2</td>
<td>24.9 ± 4.1a</td>
</tr>
<tr>
<td>Araneae</td>
<td>1.7 ± 0.3b</td>
<td>3.3 ± 0.4a</td>
<td>3.2 ± 0.6a</td>
<td>2.7 ± 0.4ab</td>
<td>2.5 ± 0.3a</td>
</tr>
<tr>
<td>Staphylinidae</td>
<td>0.7 ± 0.2</td>
<td>0.4 ± 0.1</td>
<td>0.3 ± 0.1</td>
<td>0.3 ± 0.1</td>
<td>0.6 ± 0.1a</td>
</tr>
<tr>
<td>Carabidae</td>
<td>0.3 ± 0.1</td>
<td>0.3 ± 0.1</td>
<td>0.1 ± 0.1</td>
<td>0.2 ± 0.1</td>
<td>0.4 ± 0.1a</td>
</tr>
<tr>
<td>Formicidae</td>
<td>15.8 ± 3.7</td>
<td>10.8 ± 1.8</td>
<td>7.7 ± 1.2</td>
<td>10.4 ± 1.8</td>
<td>13.3 ± 2.1a</td>
</tr>
</tbody>
</table>

<sup>a</sup>Values are mean number of arthropods per day.

<sup>b</sup>There was no significant main effect of groundcover on any arthropod group.

<sup>c</sup>Means followed by lower case letters indicate a significant groundcover by pot position interaction.

<sup>d</sup>Means followed by the same letter within a row are not significantly different ($P = 0.05$).
Table 2a. ANOVA statistics for the abundance of arthropods within wood mulch or weed cloth groundcover habitats for 6 August 2003.

<table>
<thead>
<tr>
<th>Arthropod Group</th>
<th>Effect</th>
<th>$F$</th>
<th>ndf, ddf</th>
<th>$P$</th>
</tr>
</thead>
<tbody>
<tr>
<td>Collembola</td>
<td>Groundcover</td>
<td>0.3</td>
<td>1,32.6</td>
<td>0.60</td>
</tr>
<tr>
<td></td>
<td>Pot Position</td>
<td>0.2</td>
<td>1,32.6</td>
<td>0.64</td>
</tr>
<tr>
<td></td>
<td>Groundcover × pot position</td>
<td>1.4</td>
<td>1,32.6</td>
<td>0.25</td>
</tr>
<tr>
<td>Araneae</td>
<td>Groundcover</td>
<td>1.7</td>
<td>1,33</td>
<td>0.21</td>
</tr>
<tr>
<td></td>
<td>Pot Position</td>
<td>3.2</td>
<td>1,33</td>
<td>0.08</td>
</tr>
<tr>
<td></td>
<td>Groundcover × pot position</td>
<td>7.4</td>
<td>1,33</td>
<td>0.01</td>
</tr>
</tbody>
</table>

Table 2b. Statistics for the abundance of arthropods within wood mulch or weed cloth groundcover habitats for 6 August 2003. Analysis was done using the Sheirer-Ray-Hare non-parametric method.

<table>
<thead>
<tr>
<th>Arthropod Group</th>
<th>Effect</th>
<th>$H$</th>
<th>ndf, ddf</th>
<th>$P$</th>
</tr>
</thead>
<tbody>
<tr>
<td>Staphylinidae</td>
<td>Groundcover</td>
<td>5.0</td>
<td>1,44</td>
<td>&lt;0.05</td>
</tr>
<tr>
<td></td>
<td>Pot Position</td>
<td>0.2</td>
<td>1,44</td>
<td>&gt;0.05</td>
</tr>
<tr>
<td></td>
<td>Groundcover × pot position</td>
<td>0.2</td>
<td>1,44</td>
<td>&gt;0.05</td>
</tr>
<tr>
<td>Carabidae</td>
<td>Groundcover</td>
<td>11.2</td>
<td>1,44</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td></td>
<td>Pot Position</td>
<td>2.3</td>
<td>1,44</td>
<td>&gt;0.05</td>
</tr>
<tr>
<td></td>
<td>Groundcover × pot position</td>
<td>0.1</td>
<td>1,44</td>
<td>&gt;0.05</td>
</tr>
<tr>
<td>Formicidae</td>
<td>Groundcover</td>
<td>2.7</td>
<td>1,44</td>
<td>&gt;0.05</td>
</tr>
<tr>
<td></td>
<td>Pot Position</td>
<td>0.02</td>
<td>1,44</td>
<td>&gt;0.05</td>
</tr>
<tr>
<td></td>
<td>Groundcover × pot position</td>
<td>1.6</td>
<td>1,44</td>
<td>&gt;0.05</td>
</tr>
</tbody>
</table>
Table 3. Mean ± SEM abundance of the most common arthropod groups sampled from the ground within wood mulch and weed cloth groundcover habitats summed over 3 dates in 2004.

<table>
<thead>
<tr>
<th>Arthropod Group</th>
<th>Number of Arthropods</th>
<th>Wood Mulch</th>
<th>Weed Cloth</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>Above Ground</td>
<td>Pot-in-pot</td>
</tr>
<tr>
<td>Collembola</td>
<td>23.8 ± 1.5</td>
<td>27.9 ± 4.1</td>
<td>21.5 ± 3.1</td>
</tr>
<tr>
<td>Araneae</td>
<td>2.7 ± 0.3</td>
<td>4.5 ± 0.7</td>
<td>2.6 ± 0.4</td>
</tr>
<tr>
<td>Staphylinidae</td>
<td>3.2 ± 0.4</td>
<td>2.9 ± 1.1</td>
<td>2.1 ± 0.4</td>
</tr>
<tr>
<td>Carabidae</td>
<td>0.6 ± 0.07</td>
<td>0.2 ± 0.06</td>
<td>0.3 ± 0.06</td>
</tr>
<tr>
<td>Formicidae</td>
<td>7.5 ± 0.9</td>
<td>3.1 ± 0.3</td>
<td>5.9 ± 0.7</td>
</tr>
</tbody>
</table>

*a* Values are calculated arthropods per day.

*b* There were no significant interactive or main treatment effects.
Table 4. ANOVA statistics for the abundance of arthropods within wood mulch or weed cloth groundcover habitats summed over 3 dates in 2004.

<table>
<thead>
<tr>
<th>Trophic Group</th>
<th>Effect</th>
<th>$F$</th>
<th>ndf, ddf</th>
<th>$P$</th>
</tr>
</thead>
<tbody>
<tr>
<td>Collembola</td>
<td>Groundcover</td>
<td>0.04</td>
<td>1, 8</td>
<td>0.85</td>
</tr>
<tr>
<td></td>
<td>Pot Position</td>
<td>0.01</td>
<td>1, 8</td>
<td>0.93</td>
</tr>
<tr>
<td></td>
<td>Groundcover × pot position</td>
<td>0.04</td>
<td>1, 8</td>
<td>0.85</td>
</tr>
<tr>
<td>Araneae</td>
<td>Groundcover</td>
<td>0.1</td>
<td>1, 8</td>
<td>0.81</td>
</tr>
<tr>
<td></td>
<td>Pot Position</td>
<td>0.6</td>
<td>1, 8</td>
<td>0.48</td>
</tr>
<tr>
<td></td>
<td>Groundcover × pot position</td>
<td>0.04</td>
<td>1, 8</td>
<td>0.84</td>
</tr>
<tr>
<td>Staphylinidae</td>
<td>Groundcover</td>
<td>0.1</td>
<td>1, 8</td>
<td>0.83</td>
</tr>
<tr>
<td></td>
<td>Pot Position</td>
<td>0.01</td>
<td>1, 8</td>
<td>0.91</td>
</tr>
<tr>
<td></td>
<td>Groundcover × pot position</td>
<td>0.2</td>
<td>1, 8</td>
<td>0.69</td>
</tr>
<tr>
<td>Carabidae</td>
<td>Groundcover</td>
<td>0.3</td>
<td>1, 8</td>
<td>0.61</td>
</tr>
<tr>
<td></td>
<td>Pot Position</td>
<td>1.7</td>
<td>1, 8</td>
<td>0.23</td>
</tr>
<tr>
<td></td>
<td>Groundcover × pot position</td>
<td>1.3</td>
<td>1, 8</td>
<td>0.29</td>
</tr>
<tr>
<td>Formicidae</td>
<td>Groundcover</td>
<td>0.1</td>
<td>1, 8</td>
<td>0.81</td>
</tr>
<tr>
<td></td>
<td>Pot Position</td>
<td>1.9</td>
<td>1, 8</td>
<td>0.21</td>
</tr>
<tr>
<td></td>
<td>Groundcover × pot position</td>
<td>1.2</td>
<td>1, 8</td>
<td>0.30</td>
</tr>
</tbody>
</table>
Table 5. Mean ± SEM abundance of the most common arthropod groups sampled from the soil surface within pot-above-ground and pot-in-pot plant containers in wood mulch and weed cloth groundcover habitats for 6 August 2003.

<table>
<thead>
<tr>
<th>Arthropod Group</th>
<th>Number of Arthropods</th>
<th>Wood Mulch</th>
<th>Weed Cloth</th>
<th>Above Ground</th>
<th>Pot-in-pot</th>
<th>Above Ground</th>
<th>Pot-in-pot</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>Above Ground</td>
<td>Pot-in-pot</td>
<td>Above Ground</td>
<td>Pot-in-pot</td>
<td>Above Ground</td>
<td>Pot-in-pot</td>
</tr>
<tr>
<td>Collembola</td>
<td>8.3 ± 1.8</td>
<td>159.7 ± 36.8</td>
<td>7.9 ± 2.0</td>
<td>93.1 ± 26.3</td>
<td>8.1 ± 1.3</td>
<td>126.4 ± 23.2</td>
<td></td>
</tr>
<tr>
<td>Araneae</td>
<td>0.06 ± 0.02</td>
<td>1.2 ± 0.6</td>
<td>0.1 ± 0.03</td>
<td>0.4 ± 0.1</td>
<td>0.07 ± 0.02</td>
<td>0.8 ± 0.3</td>
<td></td>
</tr>
<tr>
<td>Staphylinidae</td>
<td>0.02 ± 0.01</td>
<td>0.3 ± 0.1</td>
<td>0.1 ± 0.02</td>
<td>0.1 ± 0.04</td>
<td>0.04 ± 0.01</td>
<td>0.2 ± 0.06</td>
<td></td>
</tr>
<tr>
<td>Carabidae</td>
<td>0.03 ± 0.01</td>
<td>0.1 ± 0.04</td>
<td>0.03 ± 0.02</td>
<td>0.06 ± 0.02</td>
<td>0.03 ± 0.01</td>
<td>0.1 ± 0.02</td>
<td></td>
</tr>
<tr>
<td>Formicidae</td>
<td>0.7 ± 0.2</td>
<td>2.1 ± 0.5</td>
<td>0.8 ± 0.6</td>
<td>2.6 ± 0.9</td>
<td>0.8 ± 0.3</td>
<td>2.3 ± 0.5</td>
<td></td>
</tr>
</tbody>
</table>

* Values are mean number of arthropods per day
* All trophic groups examined were significantly more abundant within pot-in-pot plant containers compared to above ground.
Table 6a. ANOVA statistics for the abundance of arthropods sampled from the soil surface within containers from both wood mulch or weed cloth groundcover habitats and pot-in-pot and pot-above-ground containers for 6 August 2003.

<table>
<thead>
<tr>
<th>Arthropod Group</th>
<th>Effect</th>
<th>$F$</th>
<th>ndf, ddf</th>
<th>$P$</th>
</tr>
</thead>
<tbody>
<tr>
<td>Collembola</td>
<td>Groundcover</td>
<td>2.5</td>
<td>1, 33</td>
<td>0.13</td>
</tr>
<tr>
<td></td>
<td>Pot Position</td>
<td>97.9</td>
<td>1, 33</td>
<td>&lt;0.01</td>
</tr>
<tr>
<td></td>
<td>Groundcover × pot position</td>
<td>1.2</td>
<td>1, 33</td>
<td>0.28</td>
</tr>
</tbody>
</table>

Table 6b. Statistics for the abundance of arthropods within wood mulch or weed cloth groundcover habitats for 6 August 2003. Analysis was done using the Sheirer-Ray-Hare non-parametric method.

<table>
<thead>
<tr>
<th>Arthropod Group</th>
<th>Effect</th>
<th>$H$</th>
<th>ndf, ddf</th>
<th>$P$</th>
</tr>
</thead>
<tbody>
<tr>
<td>Araneae</td>
<td>Groundcover</td>
<td>0.4</td>
<td>1, 44</td>
<td>&gt; 0.05</td>
</tr>
<tr>
<td></td>
<td>Pot Position</td>
<td>24.4</td>
<td>1, 44</td>
<td>&lt; 0.01</td>
</tr>
<tr>
<td></td>
<td>Groundcover × pot position</td>
<td>1.9</td>
<td>1, 44</td>
<td>&gt; 0.05</td>
</tr>
<tr>
<td>Staphylinidae</td>
<td>Groundcover</td>
<td>0.04</td>
<td>1, 44</td>
<td>&gt; 0.05</td>
</tr>
<tr>
<td></td>
<td>Pot Position</td>
<td>7.6</td>
<td>1, 44</td>
<td>&lt; 0.01</td>
</tr>
<tr>
<td></td>
<td>Groundcover × pot position</td>
<td>1.1</td>
<td>1, 44</td>
<td>&gt; 0.05</td>
</tr>
<tr>
<td>Carabidae</td>
<td>Groundcover</td>
<td>1.7</td>
<td>1, 44</td>
<td>&gt; 0.05</td>
</tr>
<tr>
<td></td>
<td>Pot Position</td>
<td>8.9</td>
<td>1, 44</td>
<td>&lt; 0.01</td>
</tr>
<tr>
<td></td>
<td>Groundcover × pot position</td>
<td>0.9</td>
<td>1, 44</td>
<td>&gt; 0.05</td>
</tr>
<tr>
<td>Formicidae</td>
<td>Groundcover</td>
<td>1.6</td>
<td>1, 44</td>
<td>&gt; 0.05</td>
</tr>
<tr>
<td></td>
<td>Pot Position</td>
<td>17.2</td>
<td>1, 44</td>
<td>&lt; 0.01</td>
</tr>
<tr>
<td></td>
<td>Groundcover × pot position</td>
<td>0.6</td>
<td>1, 44</td>
<td>&gt; 0.05</td>
</tr>
</tbody>
</table>
Table 7. Mean ± SEM abundance of the most common arthropod guilds sampled from the soil surface within plant containers in wood mulch and weed cloth groundcover habitats and pot-above-ground and pot-in-pot containers summed over 3 dates in 2004.

<table>
<thead>
<tr>
<th>Arthropod Group</th>
<th>Number of Arthropods</th>
<th>Wood Mulch</th>
<th>Weed Cloth</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>Above Ground</td>
<td>Pot-in-pot</td>
</tr>
<tr>
<td>Collembola</td>
<td></td>
<td>23.3 ± 9.9</td>
<td>30.4 ± 11.1</td>
</tr>
<tr>
<td>Araneae</td>
<td></td>
<td>0.4 ± 0.1</td>
<td>0.5 ± 0.1</td>
</tr>
<tr>
<td>Staphylinidae</td>
<td></td>
<td>0.1 ± 0.04</td>
<td>0.2 ± 0.03</td>
</tr>
<tr>
<td>Carabidae</td>
<td></td>
<td>0.2 ± 0.05</td>
<td>0.1 ± 0.02</td>
</tr>
<tr>
<td>Formicidae</td>
<td></td>
<td>2.2 ± 0.6</td>
<td>2.4 ± 0.9</td>
</tr>
</tbody>
</table>

\(^a\) Values are mean number of arthropods per day  
\(^b\) There was no significant main effect of groundcover on any arthropod group.  
\(^c\) Means without letters indicate no significant groundcover by pot position interaction or significant groundcover by pot position interaction.  
\(^d\) Means followed by the same letter within a row are not significantly different \((P = 0.05)\).
Table 8. ANOVA statistics for the abundance of arthropods sampled from the soil surface within containers from both wood mulch and weed cloth groundcover habitats and pot-in-pot and pot-above-ground containers for Summer 2004.

<table>
<thead>
<tr>
<th>Trophic Group</th>
<th>Effect</th>
<th>$F$</th>
<th>ndf, ddf</th>
<th>$P$</th>
</tr>
</thead>
<tbody>
<tr>
<td>Collembola</td>
<td>Groundcover</td>
<td>0.02</td>
<td>1, 8</td>
<td>0.89</td>
</tr>
<tr>
<td></td>
<td>Pot Position</td>
<td>10.7</td>
<td>1, 8</td>
<td>0.01</td>
</tr>
<tr>
<td></td>
<td>Groundcover × pot position</td>
<td>0.1</td>
<td>1, 8</td>
<td>0.76</td>
</tr>
<tr>
<td>Araneae</td>
<td>Groundcover</td>
<td>0.1</td>
<td>1, 8</td>
<td>0.77</td>
</tr>
<tr>
<td></td>
<td>Pot Position</td>
<td>10.9</td>
<td>1, 8</td>
<td>0.01</td>
</tr>
<tr>
<td></td>
<td>Groundcover × pot position</td>
<td>0.3</td>
<td>1, 8</td>
<td>0.61</td>
</tr>
<tr>
<td>Staphylinidae</td>
<td>Groundcover</td>
<td>0.01</td>
<td>1, 139</td>
<td>0.91</td>
</tr>
<tr>
<td></td>
<td>Pot Position</td>
<td>18.7</td>
<td>1, 139</td>
<td>&lt; 0.01</td>
</tr>
<tr>
<td></td>
<td>Groundcover × pot position</td>
<td>1.4</td>
<td>1, 139</td>
<td>0.23</td>
</tr>
<tr>
<td>Carabidae</td>
<td>Groundcover</td>
<td>5.6</td>
<td>1, 8</td>
<td>0.05</td>
</tr>
<tr>
<td></td>
<td>Pot Position</td>
<td>13.0</td>
<td>1, 8</td>
<td>0.01</td>
</tr>
<tr>
<td></td>
<td>Groundcover × pot position</td>
<td>2.5</td>
<td>1, 8</td>
<td>0.16</td>
</tr>
<tr>
<td>Formicidae</td>
<td>Groundcover</td>
<td>0.1</td>
<td>1, 8</td>
<td>0.81</td>
</tr>
<tr>
<td></td>
<td>Pot Position</td>
<td>2.9</td>
<td>1, 8</td>
<td>0.13</td>
</tr>
<tr>
<td></td>
<td>Groundcover × pot position</td>
<td>0.01</td>
<td>1, 8</td>
<td>0.91</td>
</tr>
</tbody>
</table>
Table 9. Mean $\pm$ SEM abundance of the most common arthropod groups sampled from ground pitfall traps in plots treated with insecticide and not treated summed over 3 dates in 2004.

<table>
<thead>
<tr>
<th>Arthropod group</th>
<th>Treatment$^a$</th>
<th>Insecticide</th>
<th>No Insecticide</th>
</tr>
</thead>
<tbody>
<tr>
<td>Collembola</td>
<td>20.4 ± 3.5a</td>
<td>24.8 ± 2.6a</td>
<td></td>
</tr>
<tr>
<td>Araneae</td>
<td>2.9 ± 0.5a</td>
<td>3.1 ± 0.4a</td>
<td></td>
</tr>
<tr>
<td>Staphylinidae</td>
<td>2.5 ± 0.6a</td>
<td>3.1 ± 0.5a</td>
<td></td>
</tr>
<tr>
<td>Carabidae</td>
<td>0.4 ± 0.1a</td>
<td>0.3 ± 0.1a</td>
<td></td>
</tr>
<tr>
<td>Formicidae</td>
<td>5.2 ± 0.9a</td>
<td>5.6 ± 0.4a</td>
<td></td>
</tr>
</tbody>
</table>

$^a$ Means followed by the same letter within a row are not significant at $P = 0.05$. 
Table 10. ANOVA statistics for the abundance of arthropods sampled from ground pitfall traps in plots treated and untreated with insecticide for summer 2004.

<table>
<thead>
<tr>
<th>Trophic group</th>
<th>Contrast</th>
<th>F</th>
<th>ndf, ddf</th>
<th>P</th>
</tr>
</thead>
<tbody>
<tr>
<td>Collembola</td>
<td>Insecticide vs No Insecticide</td>
<td>0.2</td>
<td>1, 8.0</td>
<td>0.64</td>
</tr>
<tr>
<td>Araneae</td>
<td>Insecticide vs No Insecticide</td>
<td>0.1</td>
<td>1, 8.0</td>
<td>0.83</td>
</tr>
<tr>
<td>Staphylinidae</td>
<td>Insecticide vs No Insecticide</td>
<td>0.03</td>
<td>1, 8.1</td>
<td>0.87</td>
</tr>
<tr>
<td>Carabidae</td>
<td>Insecticide vs No Insecticide</td>
<td>0.2</td>
<td>1, 7.8</td>
<td>0.71</td>
</tr>
<tr>
<td>Formicidae</td>
<td>Insecticide vs No Insecticide</td>
<td>0.1</td>
<td>1, 8.3</td>
<td>0.75</td>
</tr>
</tbody>
</table>
Table 11. Mean ± SEM abundance of the most common arthropod groups sampled from within plant containers in plots treated and untreated with insecticide for 3 dates in summer 2004.

<table>
<thead>
<tr>
<th>Trophic Group</th>
<th>Treatmenta</th>
<th>Treatmenta</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Insecticide</td>
<td>No Insecticide</td>
</tr>
<tr>
<td>Collembola</td>
<td>20.28 ± 8.1a</td>
<td>34.7 ± 6.7a</td>
</tr>
<tr>
<td>Araneae</td>
<td>1.3 ± 0.3a</td>
<td>0.8 ± 0.1a</td>
</tr>
<tr>
<td>Staphylinidae</td>
<td>0.3 ± 0.1a</td>
<td>0.2 ± 0.05a</td>
</tr>
<tr>
<td>Carabidae</td>
<td>0.4 ± 0.1a</td>
<td>0.2 ± 0.04b</td>
</tr>
<tr>
<td>Formicidae</td>
<td>3.6 ± 0.7a</td>
<td>3.2 ± 0.6a</td>
</tr>
</tbody>
</table>

a Different letters in a row denote a significant difference at
Table 12. ANOVA statistics for the abundance of arthropods sampled from within containers from both treated (Insecticide) with or untreated (No Insecticide) for 3 dates in summer 2004.

<table>
<thead>
<tr>
<th>Trophic Group</th>
<th>Contrast</th>
<th>$F$</th>
<th>ndf, ddf</th>
<th>$P$</th>
</tr>
</thead>
<tbody>
<tr>
<td>Collembola</td>
<td>Insecticide vs No Insecticide</td>
<td>3.5</td>
<td>1, 9.4</td>
<td>0.09</td>
</tr>
<tr>
<td>Araneae</td>
<td>Insecticide vs No Insecticide</td>
<td>2.5</td>
<td>1, 9.1</td>
<td>0.15</td>
</tr>
<tr>
<td>Staphylinidae</td>
<td>Insecticide vs No Insecticide</td>
<td>0.6</td>
<td>1, 7.8</td>
<td>0.48</td>
</tr>
<tr>
<td>Carabidae</td>
<td>Insecticide vs No Insecticide</td>
<td>6.3</td>
<td>1, 104</td>
<td>&lt; 0.01</td>
</tr>
<tr>
<td>Formicidae</td>
<td>Insecticide vs No Insecticide</td>
<td>0.2</td>
<td>1, 7.7</td>
<td>0.69</td>
</tr>
</tbody>
</table>
Table 13. Chi – square statistics for the survival of *S. exigua* comparing the three main treatments of groundcover, pot position, and herbivore location during 3 separate field trials in summer 2003.

<table>
<thead>
<tr>
<th>Test Comparison</th>
<th>Trial 1</th>
<th></th>
<th>Trial 2</th>
<th></th>
<th>Trial 3</th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>$\chi^2$</td>
<td>$P$</td>
<td>$\chi^2$</td>
<td>$P$</td>
<td>$\chi^2$</td>
<td>$P$</td>
</tr>
<tr>
<td>Wood Mulch*Weed Cloth</td>
<td>6.4</td>
<td>0.01</td>
<td>2.3</td>
<td>0.13</td>
<td>1.5</td>
<td>0.22</td>
</tr>
<tr>
<td>Pot-in-pot *Pot-above-ground</td>
<td>1.6</td>
<td>0.21</td>
<td>1.2</td>
<td>0.28</td>
<td>0</td>
<td>1</td>
</tr>
<tr>
<td>Soil*Foliar</td>
<td>14.4</td>
<td>&lt; 0.01</td>
<td>2.3</td>
<td>0.13</td>
<td>10.8</td>
<td>&lt; 0.01</td>
</tr>
</tbody>
</table>

*a* For each treatment tactic, n = 48.
Fig. 1. Layout of treatment plot (either weed cloth or wood mulch groundcover) and pitfall traps.
Fig. 2. Mean abundance of spiders pooled (a) and the spider families: Salticidae (b), Aranaeidae (c), and Thomiscidae (d) beat sampled from foliage during the day in September 2004.
Fig. 3. Mean abundance of spiders pooled (a) and the spider families: Salticidae (b), Thomiscidae (c), and Anyphaeiniidae (d) beat sampled from azalea foliage during the night in October 2004.
Fig. 4. *Stephanitis pyrioides* azalea lace bug (ALB) survival over the season in 2004 with two groundcover types (wood mulch and weed cloth) and two positions (pot-in-pot and pot-above-ground).
Fig. 5. Mean proportion of *Agrotis ipsilon*, black cutworm (BCW) mortality for day (a) and night (b) in September 2004. Values are pooled across ground covers of wood mulch and weed cloth.
Fig. 6. The mean number of observed predation events during corresponding *Agrotis ipsilon* survival trials during the day (a) and at night (b). Predation event frequencies are total number of predation events observed over the length of each trial. Values are pooled across ground cover.
Fig. 7. The mean number of acts of predation observed by predator family for both day (a) and night (b) that correspond to the *Agrotis ipsilon* survival trials. Values are summed over the course of the respective trial.
Fig. 8. Mean number of arthropod families observed during the day and night *Agrotis ipsilon* survival trials consuming larvae in September 2004.
Chapter 3

Augmentative Biological Control: Predatory Mite Releases to Suppress Southern Red Mite, *Oligonychus illicis* (Acari: Tetranychidae)

INTRODUCTION

Production ornamental nurseries like other managed ecosystems incur frequent pest outbreaks that result in serious economic and aesthetic damage to plants (Sadof and Raupp 1996, Klingeman et al. 2000). Among the myriad of tactics available to manage pests, the most commonly used is synthetic pesticides (Hudson et al. 1996). As ornamental plant production increases in size and cash value in the United States, the use of synthetic pesticides to maintain high quality, aesthetically attractive plants will increase as well (Raupp 1995). Societal concerns regarding the environment and human health and safety, as well as increased governmental regulations have resulted in greater emphasis for alternative, less toxic pest management tactics (Garber et al. 1996).

Biological control has a long history as an alternative to manage pests, and there is increased interest in the use biological control. In many natural habitats, an assemblage of endemic natural enemies exists that can effectively suppress pest populations below economically damaging levels. However, in managed habitats, such as production nurseries and agricultural crops, golf courses, and landscapes, plant management practices often result in a decrease in natural enemy populations (Raupp et al. 1992, Hanks and Denno 1993, Tooker and Hanks 2000, Shrewsbury *in press*).

Without existing natural enemy populations in place, pest populations may grow exponentially and cause economic damage. In systems where there is a reduced or non-existent natural enemy assemblage, a pest management alternative for growers to
consider is augmentative biological control. Natural enemies used for augmentative control are mass reared in insectaries and released into a habitat with pest problems (Raupp et al. 1993, Van Driesche and Bellows 1996). Predators are available for purchase by both private consumers and commercial producers.

Most previous work that has looked at augmentative biological control against pest insects and mites has been done in very controlled environments such as greenhouses (Boys and Burbutis 1972, Hamlen and Lindquist 1981, Smith et al. 1993, Cashion et al. 1994, Pratt and Croft 1998, 2000, Schausberger and Walzer 2001, Opit et al. 2004) or laboratories (Boyne and Hain 1983), and these studies have documented varying levels of success. To a lesser extent, augmentative biological control studies have been conducted in less controlled environments, such as the landscape or production nurseries, also with varying degrees of success (Pratt and Croft 1998, 2000, Skirvin and De Courcy Williams 1999, Shrewsbury and Smith-Fiola 2000, Pratt et al. 2002, Shrewsbury and Hardin 2003). Only a few of these studies examined the role of phytoseiid mites to control ornamental pest mites (Pratt and Croft 1998, 2000, Skirvin and De Courcy Williams 1999, Pratt et al. 2002, Shrewsbury and Hardin 2003). Though the number of studies in recent years has increased my knowledge of the interactions of phytoseiid mites and phytophagous mites, there is more to understand in order to most effectively utilize augmentative releases of predatory mites as a control tactic.

Phytoseiid mites are one of the more studied groups of predatory mites, known for their success in controlling spider mites, other mites, and insects (e.g. thrips) (Gerson et al. 2003). Many families of phytoseiid mites are available from commercial biological control suppliers (Hunter 1997). McMurtry and Croft (1997) categorized phytoseiid
mites into four life style types based on life history and physical characteristics. Type I are specialized predators of heavily webbing spider mites, *Tetranychus* spp, while Type II are predators that feed on mites in the family Tetranychidae as well as on pollen and plant exudates. Type III mites are generalists that prefer to prey upon organisms other than spider mites, such as tarsonemid mites and thrips. Lastly, type IV mites are generalists that appear to develop and reproduce best on pollen (McMurtry and Croft 1997).

Phytoseiid mites, classified as either type I or type II, are used frequently in augmentative biological control programs (McMurtry and Croft 1997). A review of the literature indicated the appropriate predator species for this augmentative control study would be type II species, used often in previous studies. Predators classified as type I were not selected in this study despite reports from the literature that these predators, such as *Phytoseiulus persimilis* Athias-Henriot possess a high numerical response and quick generation time, because of the knowledge that these predators feed almost exclusively on *Tetranychus* species, and rarely upon *Oligonychus* species (McMurtry and Croft 1997, Gerson et al. 2003). Conversely, type II predators have a broader diet in which they are selective for any member of the Tetranychidae family. Many studies have documented the use of type II phytoseiids as predators, especially *Galendromous* and *Neoseiulus*, on *Oligonychus* species (McMurtry and Croft 1997, Croft et al. 1998, Pratt and Croft 2000).

Until recently, most augmentative biological control studies used single species releases of phytoseiid predatory mites to suppress phytophagous mite species. A study by Schausberger and Walzer (2001) showed that combined releases of predatory mites showed promise as a long-term biological control of spider mites. A specialist predator,
Phytoseiulus persimilis Athias-Henriot, and a generalist predator, Neoseiulus californicus McGregor, were released individually and together. When released in combination with N. californicus, P. persimilis population numbers reached higher densities than when released alone. Also, N. californicus populations grew and declined more gradually in the treatments combined with P. persimilis. These patterns were attributed to inter- and intraspecific competition. To control spider mites in perennial crops, Schausberger and Walzer (2001) suggested releases of P. persimilis alone for immediate suppression, and combination and sequential releases of N. californicus and P. persimilis for long-term control.

However, the controversy surrounding the effectiveness of multiple species is apparent in the literature. Unlike Schausberger and Walzer (2001), Shrewsbury and Hardin (2003) released G. occidentalis and N. fallacis in a 1:1 ratio on Juniperus chinensis ‘Sargentii’ A. Henry to control O. ununguis on junipers in outdoor simulated nursery environments and determined that control of spruce spider mite, Oligonychus ununguis Jacobi was poor overall and did not significantly differ from plants that had single releases of each predatory mite species. They suggest that high initial densities of O. ununguis may be responsible for this failure. Studies by Shrewsbury and Hardin (2003) suggest predator to prey ratios between 1:25 and 1:50 should provide control of O. ununguis. Opit et al (2004) found on greenhouse grown geraniums that predator: pray ratios of 1: 4 and 1: 20 significantly reduced pest T. urticae densities as well as damage to geranium foliage. Both of these studies demonstrate the importance of determining optimal predator:prey ratios.
In this study, two species of predatory mites were selected, *Galendromus occidentalis* and *Neoseiulus fallacis* (Acari: Phytoseiidae) and released in combination and at varying predator: prey ratios (1:25 and 1:50) to manage southern red mite, *Oligonychus illicis* McGregor (Acari: Tetranychidae) on holly, *Ilex meserveae* cv. Blue Princess, in a production nursery. *Oligonychus illicis* is a key pest on a number of horticultural crops, especially broadleaf evergreens, that include azaleas, camellias, and Japanese hollies (Day 2003). *Oligonychus illicis* overwinter as eggs glued to the lower leaf surface of their host plant. They are considered cool season mites and are known to have increased activity in the spring and fall months and reduced activity during summer and winter. The feeding by *O. illicis* results in mesophyll collapse and therefore, characteristic ‘stippling’ damage on infested leaves. Sufficient damage to leaves can result in premature drop and, if left unmanaged, the plant may ultimately die. This damage results in loss of aesthetic and economic value to infested broadleaf evergreens. Current methods to manage *O. illicis* are synthetic chemicals as well as dormant and horticultural oils (Shetlar and Herms 1997, Davidson and Raupp 1999). However, pesticides vary in their effectiveness and toxicity. There have not been any reports of predatory mites as augmentative biological control for *O. illicis*.

*Galendromus occidentalis* and *N. fallacis* were selected for several reasons. A species in the same genus, *Oligonychus ununguis* Jacobi, was successfully controlled with the introduction of *N. fallacis* in both natural and manipulated systems (Boyne and Hain 1983; Kramer and Hain 1989; Pratt et al. 1999, 2002; Pratt and Croft 2000a). Environmental conditions, such as temperature and relative humidity that could affect predator activity could not be predicted at the initiation of this study. Therefore upon
further literature review, I selected predators that varied in their temperature and humidity requirements for releases of a combination of two species. When Pratt and Croft (2000b) examined the life history traits of phytoseiids, they found that \textit{G. occidentalis} tolerates relative humidity levels as low as 28\%, whereas others reported that \textit{N. fallacis} performs best with a relative humidity >70\% (Boyne and Hain 1983, Kramer and Haine 1989, Mangini and Hain 1991, Pratt and Croft 2000b). In addition, we wanted to vary other life history traits, such as feeding preference. \textit{Galendromus occidentalis} is classified as having no preference for prey eggs or larvae, and \textit{N. fallacis} is known to prefer prey eggs to larvae (Schausberger and Croft 2000 a,b, Blackwood et al. 2001, Gerson et al. 2003). Other benefits of \textit{N. fallacis} are that higher densities overwinter on evergreen shrubs (Pratt and Croft 2000c), such as hollies, and that they have a relatively high intrinsic rate of increase (McMurtry and Croft 1997, reviewed by Gerson 2003). In addition, McMurtry and Croft (1997) have shown that type II phytoseiids are highly adapted to disturbed habitats.

In this study, the overall objective was to ascertain the efficacy of augmentative releases of predatory mites and compare it to the conventional method, chemical control. In addition, we wanted to examine the difference in the control of \textit{O. illicis} using varying predator mite: prey ratios. Lastly, we wanted to compare the damage incurred over the season on \textit{Ilex} leaves from the feeding by \textit{O. illicis} on plants receiving the two predator: prey ratios and chemical control measures.

\textbf{METHODS}

\textbf{Study System and Experimental Design}. Studies were conducted in a nursery in Adamstown, Maryland in 2003 and 2004. Study plants were hollies, \textit{Ilex cv. ’Blue
Princess’ that had been planted two years prior to the study. Plants were lined out at 1.2 meter centers with 1.2 meters of turf between rows. Plants were not fertilized during the experiment, and irrigation occurred via drip irrigation as needed or by rain. Hollies had a natural infestation of southern red mite, *Oligonychus illicis* McGregor (Acari: Tetranychidae).

The study was arranged as a randomized complete block design with ten replicates; each replicate contained three treatments: control, miticide, and predatory mite releases. Each treatment plot contained ten plants. For both the control and miticide treatments, two plants from each plot of ten were selected randomly to receive the treatment and were then considered the study plants used in sampling. For the predatory mite release, there were two release rates examined. Four plants were selected at random. Two plants received a predatory mite to prey ratio of 1:25; the other two received a ratio of 1:50.

**Predatory mite treatments.** Two species of predatory mites, *Neoseiulus fallacis* Garman and *Galendromus occidentalis* Nesbitt (Acari: Phytoseiidae), were used in these studies. For the two predatory mite release rate treatments examined, a 1:1 ratio of *N. fallacis* and *G. occidentalis* was used.

To determine the number of predatory mites needed for release to obtain release ratios, the number of *O. illicis* eggs and active stages present on each plant was quantified using a standardized method. First, plants were assigned to 1 of 3 size classes. Size 1 plants were approximately 53 cm tall and 43.2 cm wide. Plants assigned to size 2 were a little larger, 66 cm tall and 50.8 cm wide. The largest plants belonged to category 3, 86.4 cm tall and 63.5 cm wide. After a size category had been assigned to each plant, two
plants within each category were randomly selected, the total number of leaves on each plant was counted, and the average number of leaves per plant per size class was calculated. Ten leaves were removed from plants, five each from sides directly opposite on the plant. Leaves were selected from the top third of the plant and from areas of the branch behind the newest growth (1 year old growth). A preliminary study found this is where *O. illicis* were most commonly found on the *Ilex* (R. Waterworth, unpublished data). The number of viable eggs and active stages was counted on each leaf. The mean number of each life stage per leaf was calculated from the ten leaves. The mean was then multiplied by the total number of leaves on a plant. This provided an estimation of the total number of *O. illicis* eggs and active stages per plant. The last step in the calculation was to divide the total of *O. illicis* per plant by 25 (1:25 ratio) or 50 (1:50 ratio) to determine the number of predatory mites needed for release per plant in each treatment. The number of predator mites released on a plant ranged from 2 to 5500.

To release the appropriate number of predatory mites for each plant, mites were hand counted and released onto hollies directly with a camel hair paintbrush. In cases where the number of predatory mites to be released were too high to hand count and place, an alternative method was used. Predatory mites were shipped in vials with a sawdust material. After gently mixing the vials, 1.2 ml (=1/4 tsp) of the sawdust material was removed, placed in a petri dish, and the number of predatory mites was counted under a dissecting microscope. The average number of mites per 1.2 ml was calculated by taking three samples. The appropriate number of mites was then released on each plant with the ¼ teaspoon.
Sequential predator releases followed by *O. illicis* post counts were conducted from August 2003 through August 2004. In summer 2003, there was one count: 28 September. In summer 2004, there were four counts: 20 May, 7 July, 4 August, and 26 August. Predators were released on 17 September 2003, and 3 June, 21 July, and 12 August 2004. Post counts of *O. illicis* eggs and active stages were taken as described above (e.g. 10 leaf samples/plant). *Oligonychus illicis* data was standardized by calculating the means of eggs and active stages divided by the average leaf area from that sampling time. The number of *O. illicis* / cm² of leaf area was used in analysis. Similarly, post counts were taken on the number of predatory mites recovered at each sampling date.

The miticide, Hexygon® (hexythiox), was applied to selected plants on 12 September 2003 and 4 June 2004. An ultra fine summer oil was applied on 17 August 2004. Applications of miticides were implemented based on an increase on *O. illicis* densities on pesticide treated plants. Hexygon was applied once a year as per label restriction. Summer oil was used as an alternative method. At all three application dates, chemicals were applied at the label rate. The control was a treatment where neither predators were released or a miticide was applied. The existing population of *O. illicis* was sampled as described above at the same time as the other treatments.

**Leaf Damage Ratings.** Leaves sampled from the 26 August 2004 were evaluated for cumulative seasonal leaf damage. Two entomologists independently examined leaves from each treatment within each replicate and rated them based upon stippling damage. A scale from 0 (no damage) to 10 (heavily damaged) was used. A rating of 0 signified 0% leaf damage and 1 signified 5% leaf damage; as ratings increased by 1, the percentage
of leaf damage increased by 5%. Leaves were never damaged past 15%. The mean damage for each treatment was determined

**Predatory mite lab feeding trial.** A laboratory study was designed to determine the ability and preference of the two species of predatory mites, *N. fallacis* and *G. occidentalis* to feed on different life stages of *O. illicis*. Two trials took place: one where predators were not offered a choice between *O. illicis* life stages and one where predators were offered a choice. *Oligonychus illicis* egg and active stage survival was measured on *Ilex* leaves. All trials took place using petri dish microcosms. The leaves used in this study were undamaged and placed inside 9 cm petri dish lids on pieces of filter paper. Moisture was added to each dish by slightly moistening the filter paper with water. A smaller petri dish (5.5 cm) lid was placed over each leaf and held in place by two 5 cm binder clips (Acco, Lincolnshire, IL). For the no-choice trials, each predator species was offered only one type of prey, *O. illicis* eggs or active stages. Ten eggs or ten active stages of *O. illicis* were placed on one *Ilex* leaf. A single predator from each species was released into these microcosms. A control was also used with eggs or active stages placed on leaves but no predatory mite was released. There were five replicates of each predator type and *O. illicis* life stage combination. For the choice trials, five each of *O. illicis* eggs and active stages were placed on a single *Ilex* leaf. One predator of each species was then released into the microcosms. There was also a control in this choice trial where no predators were released into dishes. Petri dishes were incubated in a growth chamber at 23°C and 12:12 L: D. Five replicates of each predator species and control were used for this choice trial. Survival of *O. illicis* eggs and active stages was noted every 24 hours. Survival at 24 hours is reported.
Statistical Analysis. An analysis of variance (ANOVA) was used to determine the effect of the control, miticide, and natural enemy treatments on the abundance of *Oligonychus illicis* eggs and active stages as well as compare the percent leaf damage and feeding preferences of both predator species (Proc Mixed, SAS Institute 1999). The counts prior to treatment and after treatment were analyzed using Proc Mixed with a repeated measures statement (SAS Institute 1999). Additionally, planned comparisons between control, miticide, and predatory mite treatments were incorporated into the Proc Mixed procedure (SAS Institute 1999). To determine if data met the assumptions of ANOVA, homogeneity of variances and normality were examined using Proc Univariate and plotting residuals (SAS Institute 1999). Data that did not meet the assumptions of normality and homogenous variances resulted in variance partitioning, and data was then analyzed in Proc Mixed with a repeated statement. Data are presented as untransformed means ± SEM. All analysis incorporated a blocking term. Differences were considered significant at $P < 0.05$.

RESULTS

Effect of treatments on *O. illicis* abundance. There was a significant interaction of date by treatment for *O. illicis* eggs only and the total of eggs plus active stages ($F = 4.2$, df = 15, 55.6, $P < 0.01$ and $F = 3.8$, df = 15, 55.6, $P < 0.01$, respectively) (Fig. 1). While the interaction remained marginally significant for active stages only ($F = 1.8$, df = 15, 55.6, $P = 0.06$), there were significant main effects of date and treatment ($F = 14.5$, df = 5, 32, $P < 0.01$ and $F = 14.0$, df = 3, 36, $P < 0.01$, respectively) (Fig. 1). The effect of the treatments on *O. illicis* abundance varied from date to date. In general, there was a higher abundance of *O. illicis* eggs than active stages (note differences in scale of Fig.1). The
abundance of eggs and active stages of *O. illicis* remained lowest in the miticide treatment overall compared to the control and predatory mite treatments (Figs. 1a-c). The differences in the abundance of eggs alone, active stages alone, and total of eggs plus active stages between miticide and other treatments were all significant (Table 1). However, there were no significant differences in *O. illicis* abundance between the control and the combined effect of the predatory mite treatments (Table 1).

The number of predatory mites recovered following release was extremely low (range 0 – 2.4 / cm² leaf area) for all treatments and dates (Table 2).

**Leaf Damage Ratings.** Leaves taken from the final sampling date of hollies to determine *O. illicis* abundance were also used to evaluate the visible effects of the treatments on cumulative damage across the season. There was a significant effect of treatment (*F* = 7.21, df = 3, 27, *P* < 0.01), however, the predatory mite treatments (predator: prey, 1:25 and 1:50) did not differ significantly in their leaf damage compared to the control (*F* = 0, df = 1, 27, *P* = 1.0). On average, leaves from the control had a damage rating of 1.95 ± 0.27, and the 1:25 and 1:50 predatory mite treatments had ratings of 1.975 ± 0.29451 and 1.925 ± 0.22376, respectively (all ~ 10% damage) (Fig. 2). Damage to plants in the miticide treatment significantly differed from the control and predator treatments; the leaf damage rating was lower at 0.65 ± 0.21 (~ 5% damage).

**Predatory mite lab feeding trial.** In the first laboratory study, predatory mites, *N. fallacis* and *G. occidentalis*, were not given a choice between *O. illicis* life stages. A significantly lower proportion of eggs survived in the predator treatments compared to the control (*F* = 11.95, df = 2, 9, *P* < 0.01), however the percentage of eggs that survived did not differ between predator species (70.0 ± 7.1 and 52.5 ± 8.5, *G. occidentalis* and *N.*
When the predators were offered active *O. illicis*, significantly fewer survived compared to the control (*F* = 8.2, df = 2, 8, *P* = 0.01); again, however, there was no difference in the percentage survival of active stages between predator species (6.0 ± 6.0 and 4.0 ± 4.0, *G. occidentalis* and *N. fallacis*, respectively) (Fig. 3). *G. occidentalis* and *N. fallacis* both consumed eggs and active stages but both predators consumed more active stages than eggs in a no choice study.

The next experiment determined if predatory mites had a preference for either *O. illicis* eggs or active stages when offered both simultaneously. There was a significant interaction of predatory mite species by *O. illicis* life stage of (*F* = 4.62, df = 2, 19.1, *P* = 0.02). This interaction is mostly attributable to the high percentage of egg survival with *G. occidentalis* (96.0 ± 4.0) and a lower survival of eggs with *N. fallacis* (68.0 ± 10.2). For both predatory mite species, when given a choice, fewer eggs were consumed than active stages (Fig. 4).

**DISCUSSION**

The main objective of this study was to determine if augmentative releases of a combination of two predatory mite species in two predator: prey ratios could reduce pest mite densities and injury compared to conventional miticide applications and control (no treatment) plants, hollies, in a field production nursery. Overall, predator releases were not as efficacious at suppressing spider mite populations compared to conventional miticide control measures, and *O. illicis* densities on predator release plants did not differ from control plants. In addition, all treatments resulted with plants receiving spider mite injury. However, miticide treated plants had less injury than predator and control plants.
No other study has examined the use of predatory mites in the control of *O. illicis* in review of recent literature. Two previous studies have evaluated augmentative biological control to suppress another *Oligonychus* species, spruce spider mite (*O. ununguis*) density and damage on woody plants (Pratt et al. 2002, Shrewsbury and Hardin 2003). Pratt et al. (2002) used *N. fallacis* as an augmentative biological control agent and rated control of spruce spider mite on a scale of 1-4, with 1 as unacceptable and 4 being complete control. From small-scale field studies, they found *N. fallacis* provided unacceptable control with a damage rating of 1 in two of four trials. While the other two studies provided acceptable control, plants still had a damage rating of 2. In larger-scale nursery level studies, *N. fallacis* provided acceptable control in all trials but also with a plant damage rating of 2. Overall, Pratt et al (2002) found that levels of control but plants in all trials sustained damage. Similar to Pratt et al. (2002), all of my treatments resulted in foliar damage. Predatory mites at certain dates appeared to offer limited control of *O. illicis* compared to control plants, however, overall pest abundance reached unacceptable levels.

Shrewsbury and Hardin (2003) also used *N. fallacis* as well as *G. occidentalis* in augmentative releases to suppress *O. ununguis* under a simulated nursery environment. In addition to releasing each species individually, there was also a release treatment that included the combination of both species. Similar to my study, there was no significant difference in the abundance of *O. ununguis* between any of the predatory mite treatments and the control. Also similar to my study, damage ratings of plants with releases of predatory mites also did not differ significantly from the control plants. Shrewsbury and Hardin (2003) suggested that poor control by predatory mites was possibly due to an
already high *O. ununguis* population, an underestimation of the pest population via beat-sampling, and/or inadequate predator release rates (Shrewsbury and Hardin 2003). Based on their data they also suggested predator: prey ratios of 1: 25 to 1: 50 should be effective in suppressing *O. ununguis* populations. My study addressed some of these factors that Shrewsbury and Hardin suggested as reasons for lack of control. I developed a more accurate method to estimate pest abundance and also released predators in two ratios relative to their prey. However, despite these measures, I still got poor control.

Studies of spider mites with non-*Oligonychus* genera have also been conducted in outdoor environments on ornamental plants. These have found that phytoseiid mites can suppress tetranychid mites. Pratt and Croft (1998) released *N. fallacis* and found a significant reduction of *Panonychus citri* on *Skimmia japonica* grown in containers. In another study, Pratt and Croft (2000b) used three phytoseiid species, *N. fallacis*, *G. occidentalis*, and *N. californicus*, and reduced densities of *Tetranychus urticae* Koch on *Malus*, *Acer*, and *Spiraea* as well as *O. illicis* on *Rhododendron* plants. It was not clear what predator: prey release ratio was used by Pratt and Croft (2000b).

As demonstrated by the mixed success of augmentative releases of predatory mites, there are still many details that are only moderately understood and must be studied further to improve the use of biological control in outdoor environments such as production nurseries. Several factors might explain the failure of augmentative releases under conditions found in my study and possibly others. For example, predatory mite species or combinations of species may vary in their ability and/or preference to consume both pest mite eggs and active stages (Blackwood et al. 2001). In addition, the prey life stage preferred by the predator may not be the more abundant or available stage in the
system. For example in my study, more eggs than active stages of *O. illicis* were present on leaves throughout the season (Figs. 1a and b). This suggests that *N. fallacis* and *G. occidentalis* either did not consume or prefer eggs of *O. illicis*. My lab study supports this conclusion. Both species did consume eggs when only eggs were offered, however, fewer eggs were eaten when the predators were offered a choice between eggs and active stages. This was especially so for *G. occidentalis*. This is somewhat contrary to reports from previous work. Blackwood et al. (2001) showed that *N. fallacis* preferred eggs of another tetranychid mite, *Tetranychus urticae*, while *G. occidentalis* did not have any preference for either life stage. Future work should examine other predator species as potential control agents of *O. illicis* and determine the prey life stage on which they prefer to feed.

Asynchrony in life histories between prey and predator species may also hinder the ability of predators to suppress pests. *Oligonychus illicis* is classified as a cool season mite, and diapausing eggs begin to hatch as temperatures warm in late March until early May (Mague and Streu 1980). Patterns in populations have shown a dramatic decline of *O. illicis* on both hollies and azaleas in midsummer, and it is late September when the females begin to deposit eggs that populations become economically significant again (Mague and Streu 1980). Childs et al. (1984) showed in a laboratory setting that as temperatures increased there was a decrease in both net reproductive rate and the intrinsic rate of natural increase. The optimal temperature for the highest reproductive rate and intrinsic rate of increase was 26.5°C. However, the opposite phenomenon occurs for *G. occidentalis* (Gerson et al. 2003). Optimal development and reproductive rates for *G. occidentalis* occurred at 32°C. *N. fallacis* is also sensitive to temperature and daylight
and becomes active with increasing temperatures, though these thresholds are lower than with *G. occidentalis* (Gerson et al. 2003). Optimal temperatures and daylight length are important to ensure that the predators reach their highest potential as biological control agents. In Maryland, temperatures in the field by late June through August regularly exceeded 30°C which is possibly associated with the natural population decline of field *O. illicis*. With both tetranychid and phytoseiid species having opposite temporal schedules, this suggests they may limited control of *O. illicis*.

The results of this study do not correspond to what I would predict based on the reported life histories of *O. illicis* and the predators. The June release appears to have been successful relative to the control at suppressing *O. illicis* by early July, however, by early August despite another predator release, *O. illicis* populations were increasing. Populations declined in all treatments by the end of August (Fig. 1c). It appears that further studies are needed to establish *O. illicis* and predatory mite life histories.

Alternatively, two species of predatory mites were released in this system, intraguild predation might have taken place between both species or other predators that may have been present. *Galendromus occidentalis* and *N. fallacis* are both specialist predators and have traits that make them poor competitors with co-occurring phytoseiid species (reviewed by Gerson 2003). *Neoseiulus fallacis* and *G. occidentalis* are known to be easily displaced by competitors (Type III predators) that survive on non-animal diets (Gerson et al. 2003). However, there is little data on their competitiveness with each other. In addition to being poor competitors, type II predators are known to disperse from areas where sufficient food sources are not present (McMurtry and Croft 1997, Gerson et al. 2003). My study suggests either of these phenomena may be taking place in
this system. Recovery of predatory mite species released (range 0-2.4) in my study was dramatically lower than the numbers released (range 2 - 5500). It may be that *O. illicis* densities on hollies, especially active stages were too low to prevent intraguild predation or dispersal of my predators.

There has been mixed success of multiple predator species releases to control pest mites. My study found no difference in *O. illicis* abundance between the control or the predatory mite treatments in which a 1:1 ratio of 2 predator species were released. Similarly, Shrewsbury and Hardin (2003) found no significant difference in abundance of spruce spider mite between their control and the combination release of two predator species. Alternatively, releasing more than one predator species with different predatory behaviors have been found to have an impact on spider mite populations and in some cases an additive effect (McMurtry and Croft 1997, Schausberger and Walzer 2001).

Augmentative biological control may be strongly influenced by three related factors. These are timing of predator release, initial prey density, and predator release rate or predator: prey ratio (Hamlen and Lindquist 1981, Stiling 1993, Skirvin and De Courcy Williams 1999, Pratt and Croft 2000a). In my study, 2 predator: prey release ratios were examined: a high predator: prey (1:25) or a low predator: prey (1:50) ratio. These ratios were selected based on studies from Shrewsbury and Hardin (2003) that provided evidence a 1:25 predator: prey ratio should provide control. However, neither ratio suppressed *O. illicis* relative to the control (no predators). The ineffectiveness of these predator: prey ratios could possibly be the result of reasons discussed previously. However, an additional explanation could be that the ratios selected for this study were not sufficient to provide control. It is difficult to determine predator release rates, as
there is little empirical data. Release rate recommendations are available from extension fact sheets and information bulletins produced by commercial suppliers of beneficial arthropods. These recommendations however are variable and few are available for ornamental production nurseries. Studies that have examined release rates or ratios have been predominately geared for greenhouses. For greenhouse plants, recommendations from commercial producers include: two or three predators per 0.0929 m² (1 sq. foot) of foliage; 10-100 predators per plants; or one predator per five prey. Recommendations include one predator per 10 or 20 prey, or one predator per 25 prey in orchards. Another study that has more closely examined predator: prey ratios by Hamlen and Poole (1982) used Phytoseiulus macropilis (Banks) on T. urticae. Spider mite populations were suppressed at ratios of 1:5 and 1:10 while a 1:20 ratio and control resulted in significant damage to plants. A more recent study by Opit et al. (2004) examined the effect of predatory mites to suppress T. urticae and found that ratios of 1:4 and 1:20 significantly reduced pest densities and plant damage on ivy geranium in greenhouses. A ratio of 1:60 resulted in occasional significant T. urticae pest reduction, however with this ratio, predators did not protect ivy geraniums from significant damage (Opit et al. 2004). This study suggests that my ratios should have suppressed O. illicis populations. However, Opit et al. (2004) study and mine differed in several ways. The differences include indoor (greenhouse) versus outdoor setting, different predator and prey species, and predators that specialize on Tetranychus species.

Timing of predatory mite releases is essential to provide the desirable levels of control of phytophagous mites. Extension publications recommend timing releases to when spider mite populations are first seen or low. A study that enforces the idea of early
Predator releases and has shown successful results of decreasing plant damage is one by Hamlen and Lindquist (1981). They demonstrated that it was important to introduce predators at low densities of spider mites because it took one to three weeks to provide control. In my study, the initial release of predators in early June 2004 was near the peak population of *O. illicis* for the growing season. Predators that were released may have been overwhelmed by a burgeoning pest population. However, this seems unlikely since the predator number for release was estimated for each plant so that the 1:25 and 1:50 ratios remained constant treatments.

Releasing predators on a regular schedule, or sequentially, is also recommended by commercial insectaries. Sequential releases were made in my study to continually introduce predators into the system and to maintain long-term herbivore control, but control was again varied despite calendar releases.

Growers and retail nurserymen are unable to market plants that have received too much feeding damage from phytophagous arthropods. In this study, damage on control plants and those with predatory mites released averaged around 10% for each treatment. Only the miticide treatment had a lower amount of foliar damage (about 5% damage). Several studies have been done to examine the relationship between ornamental plant value and pest injury to plants to develop economic and aesthetic thresholds (Sadof and Raupp 1987, Coffelt and Schultz 1993, Sadof and Alexander 1993). Retail customers were surveyed to determine the level at which plant injury is too high and they would no longer purchase the plant. The general consensus was that once plants reached 10% injury, customers would not buy the plant. With the amount of damage on the plants in
this study, it is unlikely that these plants would be ready for retail sale, except for the miticide treated plants.

In conclusion, the predatory mite species chosen for this study may not have been appropriate, as their feeding preferences did not promote *O. illicis* suppression. Opposing life cycles of predator and prey may have also inhibited effective control where prey were active prior to establishment of the predator. Also, predators may have been susceptible to competition for resources by other mite species. The result of these predator treatments was an increase in damage to the foliage and potential unmarketability of the plants compared to a conventional miticide treatment. To increase the success of augmentative biological control in outdoor environments, the appropriate predator should be selected in order to manage the pest. Future research in this area should include the use of preference feeding trials to determine which commercially available predator species consumes eggs and active stages of *O. illicis*. Also, if multiple predator species are chosen, it is important to determine the interaction of predators to circumvent intraguild predation. Lastly, once predators are chosen, it is important to identify the predator-prey population dynamics to determine predator: prey release ratios that provide optimal control of not only *O. illicis* but other spider mites on a range of plants in outdoor environments.
Table 1. ANOVA contrasts for evaluating *Oligonychus illicis* egg and active stage abundance singularly and combined for *O. illicis* eggs and active stages for the control, miticide, and predatory mite (both 1:25 and 1:50) treatments.

<table>
<thead>
<tr>
<th>Life Stage</th>
<th>Contrast</th>
<th>$F$</th>
<th>df</th>
<th>$P$</th>
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<tr>
<td>Eggs Alone</td>
<td>Control vs. Predatory Mites</td>
<td>0.03</td>
<td>1, 36</td>
<td>0.87</td>
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<td></td>
<td>Control vs. Miticide</td>
<td>10.9</td>
<td>1, 36</td>
<td>&lt;0.01</td>
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<tr>
<td></td>
<td>Predatory Mites vs. Miticide</td>
<td>15.7</td>
<td>1, 36</td>
<td>&lt;0.01</td>
</tr>
<tr>
<td>Active Alone</td>
<td>Control vs. Predatory Mites</td>
<td>0.60</td>
<td>1, 36</td>
<td>0.44</td>
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<td>Control vs. Miticide</td>
<td>31.9</td>
<td>1, 36</td>
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<td></td>
<td>Predatory Mites vs. Miticide</td>
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<td>1, 36</td>
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<tr>
<td>Eggs + Alone</td>
<td>Control vs. Predatory Mites</td>
<td>0.01</td>
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Table 2. Mean ± SEM of predatory mites (per cm² leaf area) recovered during sampling for the abundance of *Oligonychus illicis* eggs and active stages on three dates in 2004, 7 July, 4 August, and 26 September.

<table>
<thead>
<tr>
<th>Date (2004)</th>
<th>1:25</th>
<th>1:50</th>
<th>Control</th>
<th>Miticide</th>
</tr>
</thead>
<tbody>
<tr>
<td>7 July</td>
<td>2.3 ± 0.9</td>
<td>1.8 ± 0.7</td>
<td>2.4 ± 1.0</td>
<td>0.4 ± 0.3</td>
</tr>
<tr>
<td>4 August</td>
<td>2.3 ± 0.7</td>
<td>2.0 ± 0.5</td>
<td>0.8 ± 0.3</td>
<td>1.2 ± 0.5</td>
</tr>
<tr>
<td>26 August</td>
<td>0.7 ± 0.3</td>
<td>0.2 ± 0.1</td>
<td>0.3 ± 0.2</td>
<td>0.0 ± 0.0</td>
</tr>
</tbody>
</table>
Fig. 1. The abundance (number per cm$^2$ leaf area) of *Oligonychus illicis* eggs alone (a), active stages alone (b), and eggs + active stages (c) with the four treatments: control, miticide, and predator: prey ratios of 1:25 and 1:50. The 28 August 03 is a precount of *O. illicis* abundance prior to treatments. Arrows with letters indicate: (a) predator and miticide application (Hexygon), (b) predator application, and (c) predator and oil application. Note differences in scale.
Fig. 2. Leaf damage ratings cause by *Oligonychus illicis* for *Ilex* leaves collected on 26 August 2004. Ratings equal to 1 are 1 to 5% damage, 2 are 6 to 10% damage, and 3 are 11 to 15% damage.
Fig. 3. A “no choice” lab predator preference study measuring the percentage survival of either eggs or active stages of *Oligonychus illicis* when they were offered singularly to two predatory mite species, *Galendromus occidentalis* (GO) and *Neoseiulus fallacis* (NF).
Fig. 4. A “choice” lab predator preference study to measure percentage of survival of *Oligonychus illicis* eggs and active stages, when two predators, *Galendromus occidentalis* (GO) and *Neoseiulus fallacis* (NF), were offered both life stages in the same microcosm.
REFERENCES


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