CONSERVING NATURAL ENEMIES TO CONTROL LYGUS IN

WASHINGTON STATE ALFLAFA FIELDS

By

ANN ELIZABETH JORGENSEN

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To the Faculty of Washington State University:

The members of the Committee appointed to examine the thesis of ANN ELIZABETH JORGENSEN find it satisfactory and recommend that it be accepted.

Chair

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Abstract

by Ann Elizabeth Jorgensen, M.S. Washington State University May 2005

Chair: William E. Snyder

Lygus bugs (*Lygus* spp.) are the most important insect pests in alfalfa grown for seed. Lygus are particularly damaging to seed production because they feed on the reproductive parts of the plant. Typically, Lygus bug outbreaks have been treated, in seed fields, with broad-spectrum insecticides. However, alternatives to broad-spectrum insecticides are being sought for many reasons, but most importantly because of new legislation that will limit the use of broad-spectrum insecticides. Conservation biological control is a promising alternative to insecticide use in alfalfa seed. Thus, a survey of alfalfa (*Medicago sativa*) seed and hay fields was conducted to identify insect predators that will be beneficial for controlling Lygus bugs. Samples were taken from hay fields to determine insect densities in alfalfa with low insecticide input.

The survey of alfalfa hay and seed fields was conducted during 2003 and 2004. Samples were taken from fields in Touchet, WA and Warden, WA in May, June and July

iv

of each year. Insect samples were collected using an insect suction sampler (D-vac) and pitfall traps. Lygus populations were most abundant in July, when blooms are forming on alfalfa plants. Another important pest of alfalfa is the pea aphid (*Acyrthosiphon pisum*), which also had high densities in July. The most common insect predators in July were *Hippodamia convergence, Coccinella septempunctata, Calosoma spp.*, Staphylinid beetles, *Nabis* spp., Thomisid spiders and Linyphiid spiders. Predator populations tended to be higher in hay fields than in seed fields.

In a Petri dish assay, the six most common insect predators during July were tested for Lygus and pea aphid predation. Damsel bugs and crab spiders ate the most Lygus bugs. A microcosm experiment was performed with these two predators to determine Lygus consumption in the presence and absence of pea aphids. In the presence of alternative prey, Lygus predation was reduced suggesting a positive prey-prey interaction.

In conclusion, Lygus control is most important during July when populations peak. Predator populations in hay fields suggest that there are a sufficient number of predators for biological control of Lygus, in the absence of broad-spectrum insecticide use.

v

TABLE OF CONTENTS

	Page
ACKNOWLEDGEMENTS	iii
ABSTRACT	iv
LIST OF TABLES	viii
LIST OF FIGURES	ix
<u>CHAPTER 1</u> - ALFALFA SEED PRODUCTION AND LYGUS CONTROL Washington State Alfalfa Management of Lygus bugs in Alfalfa Seed BROAD-SPECTRUM INSECTICIDES Resistance	3 4
Resurgence and Secondary Pest Outbreak Environment and Health Food Quality Protection Act Alternatives to Broad-spectrum Insecticides	5 6 7
CONSERVATION BIOLOGICAL CONTROL. Generalist Natural Enemies. Research Objectives. REFRENCES.	9 10
<u>CHAPTER 2</u> - COMPARISON OF INSECT GUILDS IN ALFALA GROWN FOR SEED, VERSUS ALFALFA GROWN FOR HAY	
INTRODUCTION METHODS AND MATERIALS Study Sites Foliar Invertebrate Sampling	19 19
Ground Invertebrate Sampling Statistics RESULTS	20 21
Lygus Pea Aphid Foliar Predators	22
Ground Predators DISCUSION REFRENCES	23

<u>CHAPTER 3</u> - PEA APHIDS MAINTAIN LEVELS OF GENERALIST FOR CONTROLLING LYGUS PESTS IN ALFALFA GE	
SEED	
INTRODUCTION	
METHODS AND MATERIALS	42
Collection of Study Organisms	
No-choice Petri Dish Experiment	
Microcosm Experiment	
Statistics	45
RESULTS	45
No-choice Petri Dish Experiment	45
Microcosm Experiment.	46
DISCUSSION	
Summary	
REFRENCES	

LIST OF TABLES

<u>Tab</u>	e Pa	age
2.1	Alfalfa field sites by year, location, type, and irrigation	.28
2.2	Logistics of pitfall samples	.29
2.3	Statistical analysis of D-vac and pitfall samples	.30
2.4	Insects identified from D-vac and pitfall samples in 2003 and 2004	.31
3.1	Microcosm statistics	.53

LIST OF FIGURES

Figure Page
2.1 Lygus, pea aphid and total predator averages for 2003 and 2004 D-vac samples3
2.2 Percentages of the most common foliar predators from D-vac samples in 2003 and 2004
2.3 Total predators averaged per sample per day in 2003 and 2004 pitfalls
2.4 Percentages of the most common ground predators from pitfalls in 2003 and 200438
3.1 Prey-prey interactions
3.2 Lygus and pea aphid consumption by generalist predators
3.3 Consumption of Lygus in the presence of alternative prey

DEDICATION

This is dedicated to my grandmother who passed during my time

here at Washington State University.

CHAPTER 1

ALFALFA SEED PRODUCTION AND LYGUS CONTROL

INTRODUCTION

Washington State Alfalfa

Alfalfa (*Medicago sativa* L.) is often referred to as the "queen of forages" because of its nutritional value and digestibility for livestock (Putman et al., 2001). Alfalfa was brought to the United States in the mid 1800's to be used as forage (Westgate 1908). Alfalfa is a perennial crop that can last for many years with proper cultivation. In Washington state, alfalfa is grown for both hay and seed production. Over 800,000 acres of alfalfa hay were harvested in Washington during 2003, with a total value of over \$340 million (WASS, 2005). Hay growers harvest their crop several times in a season; typically there are 3-4 harvests per year (Haley and Baker, 1981). The goal of hay producers is to produce a crop that is high in nutrients, in order to provide quality feed for animals. Some of the most important aspects in managing alfalfa hay are proper irrigation, weed control and disease prevention. Hay growers can tolerate higher insect pest densities than can seed growers, because hay growers can harvest their crop before pests become severe (Haley and Baker, 1981).

Alfalfa seed has been produced in Washington since 1937 (WASS, 2005). Alfalfa seed was one of the top 30 agricultural commodities for Washington State in 2003, and brought in over 13 million dollars to the state (WASS, 2005). Like alfalfa hay, alfalfa seed needs to be carefully monitored for proper irrigation, weed control and disease prevention (Rincker et al., 1987). However, there are many other factors that contribute

to a successful seed harvest, which include insect control, high levels of pollination and the proper timing of cultural practices (Mueller, 2003). Of these components insect control can be the most demanding, because of the numerous insect pests that attack alfalfa (Rincker et al., 1987).

Alfalfa often houses a diverse insect community, including both herbivores and natural enemies. There are reports of up to 591 species of insects in alfalfa (Pimentel and Wheeler, 1973). Some of the most important pests of economic significance include the Lygus bugs (Lygus hesperus Knight and Lygus elisus Van Duzee), the pea aphid (Acyrthosiphon pisum Harris), the alfalfa weevil (Hypera postica Gyllenhal), and the clover root curculio (Sitona hispidula Fabricius) (Berg and Lauderdale, 1982). Pests of lesser concern include grasshoppers, armyworms, seed chalcids, and cutworms (Berg and Laurdale, 1982; Gupta, 1979). The natural enemies commonly found in alfalfa include predatory Hemiptera, such as *Nabis*, *Geocoris*, and *Anthocoris* species (Fisher, 1982; Gupta et al., 1980). Other common natural enemies include lacewings (Neuroptera), ladybird beetles (Coleoptera: Coccinellidae), and carabid beetles (Coleoptera: Carabidae) (Fisher, 1982). Parasitoids can also be abundant in alfalfa; there are many species that attack a variety of insects (Gupta et al., 1980). Some of the most common arachnids are crab spiders (Thomisidae) (Gupta, 1977). However, of all these arthropods, the single species of greatest economic importance for alfalfa seed production are Lygus bugs (Gupta et al., 1980).

Management of Lygus Bugs in Alfalfa Seed

Lygus bugs are omnivorous hemipterans commonly associated with alfalfa, cotton and many other managed crops. They also thrive on a variety of weeds, which may facilitate early season colonization of fields (Snodgrass and Scott, 2000). Lygus are the most important pest of alfalfa seed in the Pacific Northwest because of the damage that they cause to the reproductive parts of plants, such as the petiole and buds (Gupta et al., 1980). Damage caused to the petiole and buds of alfalfa can be severe, causing shriveled reproductive parts, bud blast and seed abortion (Gupta et al., 1980). Lygus often feed directly on seeds, causing the seed to shrivel thus reducing the weight of the seed pod; if too light, the pod is not retained by harvesting equipment (Gupta et al., 1980). Since Lygus attack at a crucial stage in the development of the alfalfa plant, insecticides are used to reduce their populations (Schaber et al., 1990).

Lygus control in alfalfa seed is achieved primarily through use of broad-spectrum insecticides, most commonly organophosphates such as Monitor®, Supracide® and Dibrom® (Mueller, 1998). To control Lygus, growers often implement calendar sprays, which are insecticides applications made on a specific date, regardless of pest levels (Baird and Homan, 1991). Many alfalfa seed growers implement calendar sprays before and after pollination (Baird and Homan, 1991). Although Lygus control with organophosphates is effective, there are several problems associated with using broad-spectrum insecticides, including the eventual development of insecticide resistance, initiation of pest resurgence and secondary pest outbreaks, and negative environmental and health effects to humans (Metcalf, 1986). Concerns about health risks associated with

pesticides have resulted in new federal legislation that will restrict the use of many broad spectrum insecticides (Epstein et al., 2000).

BROAD-SPECTURM INSECTICIDES

Resistance

In 1946 the housefly became resistant to DDT (Hajek, 2004). Since DDT resistance was first recorder, there have been numerous other documented cases of resistance evolving, involving many different pesticides (Hajek, 2004; Pimental, et al., 1992). Resistance to a pesticide can occur along three main pathways: biochemically, physiologically or behaviorally, with these pathways acting individually or in concert (Georghiou, 1972). Cross resistance among pesticides has also been established between many of the broad-spectrum pesticides (Georghiou, 1972). Resistance is more prevalent among herbivores than natural enemies, and of the 400 reported cases of pesticide resistance only a fraction of those are natural enemies (Roush and McKenzie, 1987).

In alfalfa, resistance has become a problem within Lygus populations. Because of repeated insecticide applications against this pest, Lygus populations can quickly evolve resistance, often within a single season (Mueller, 2003). In addition, Lygus can develop resistance to multiple insecticides (Mueller, 2003). In one published case, Lygus bugs collected from cotton fields in Mississippi were tested for resistance to three classes of insecticides, pyrethroids, organophosphates, and cyclodienes; resistance was present for all classes (Snodgrass, 1996).

Resurgence and Secondary Pest Outbreak

In addition to resistance, resurgence and secondary pest outbreaks are often associated with heavy insecticide reliance. Both resurgence and secondary pest outbreaks are in part the result of disruption of natural enemy populations (Hardin et al., 1995). Resurgence, or the rapid recovery of pest populations to pre-spray levels, has often been shown to occur in systems where natural enemy populations are depauperate. For example, it has been shown that spider mites in apple orchards increased to outbreak levels within 2 months after spraying a broad spectrum insecticide, whereas it takes several more months for predatory mite populations to recover (Kapetanakis, Warman and Cranham, 1986); in some cases it has taken as long as 6 years for natural enemy populations to fully recover following an insecticide treatment (Hardin, et al., 1995). Due to the lag in recovery for natural enemy populations, the pest species is released from control and can then reach outbreak densities (Hajek, 2004).

Secondary pest outbreaks can also occur after insecticides disrupt a natural enemy community. Secondary pests are herbivores that only cause damage once their natural enemies are decimated by insecticides (Hardin et al., 1995). The most common secondary pests in alfalfa are aphids, mites, and thrips (Cone, 1963). However, many new alfalfa cultivars are resistant to aphids and moderate populations can be tolerated (Blodgett, 2003). A study conducted in alfalfa to determine the effects of organophosphates on mite populations revealed that the phytophagous mite *Tetranychus telarius* L. reached outbreak levels after pesticide applications targeting *Tydus* spp. (Cone, 1963); in the absence of insecticides, *T. telarius* never reached damaging densities.

Environment and Health

Environmental and health risks associated with pesticides have been an important public concern for the last 40 years. One concern is that insecticide run-off into nearby water sources might degrade water quality (Cox, 2002). For example, in Washington State there has been ongoing research into the contamination of the Columbia River by agricultural chemicals and waste (Cox, 2002). It is thought that these pollutants may be responsible for a reduction in wild salmon populations (Cox, 2002; Scholz et al., 2000). Insecticides also pose threats to humans. It is estimated there are approximately 67,000 pesticide-related poisonings, and an estimated 27 pesticide related fatalities in the U. S. each year (Pimentel et al., 1992). There have been numerous studies in Washington State assessing the exposure of children to insecticides (Curl et al., 2003; Fenske et al., 2002; Wessel et al., 2003; Wilson et al., 2003). Because of these health concerns, especially pesticides risks to children, new legislation has been enacted that will seriously limit organophosphate use (Epstein et al., 2000; Pimentel et al., 1992; Wheeler, 2002).

Food Quality Protection Act

In 1996, the Environmental Protection Agency (EPA) amended the Federal Insecticide, Fungicide, and Rodenticide Act (FIFRA), via the Food Quality Protection Act (FQPA). FQPA, as amended, now mandates that all pesticides be reviewed for their potential hazards to humans, specifically children, from contaminated food, drinking water, and home and garden use (Wheeler, 2002). Per FQPA guidelines, organophosphates are the first group of pesticides to be evaluated, followed by carbamates (Wheeler, 2002); these groups of pesticides constitute a majority of the insecticides used for insect control in alfalfa (Mueller, 2003). With the numerous reevaluations, it is considered unlikely that any new organophosphate or carbamate pesticides will be registered (Wheeler, 2002). Alfalfa seed is a successful commodity; however it is small in comparison with other crops and will most likely have difficulties registering new insecticides. With the loss of many relied-upon insecticides, new measures will need to be taken in order to control insects (Epstein et al., 2000; Wheeler, 2002).

Alternatives to Broad-spectrum Insecticides

Because of the many risks associated with the use of broad-spectrum insecticides, recent work has focused on developing selective pesticides that target specific pests, but without harm to beneficials (May et al., 2003; Wheeler, 2002). Selective pesticides are also "complementary" for incorporating effective conservation biological control into crop production (e.g., Angello et al., 2003; Koss et al., 2005). A study in New York State of control of the European red mite, in apple orchards, demonstrated that a successful control program can combine both biological control and selective insecticides (Agnello et al., 2003). In another study that examined predator and pest communities in potato fields treated with broad-spectrum, selective or organic insecticides, it was shown that fields treated with selective insecticides had predator densities similar to organic fields, and potato yields equivalent to conventionally treated fields (Koss et al., 2005). These studies illustrate that effective pest control can be attained by the use of selective insecticides in conjunction with natural enemy conservation.

CONSERVATION BIOLOGICAL CONTROL

Conserving natural enemies is a strategy of biological control where modifications are made to the environment, or existing farming practices, to enhance natural enemies and reduce pests (Ehler, 1998; Hajek, 2004). The idea of conserving natural enemies came about once the widespread use of synthetic chemical pesticides led to devastated natural enemy communities, across a broad range of cropping systems (van den Bosch and Telford, 1964). It has been widely reported that broad-spectrum insecticides have negative effects on natural enemies, both directly and indirectly (Moreby et al., 1997; Epstein et al., 2000; Michaud and Grant, 2003). Thus, one of the most effective ways to conserve natural enemies is to alter the use of pesticides either by reducing total insecticide input, strategically timing applications to minimize negative effects on natural enemies, or replacing broad spectrum insecticides with more selective chemicals (Barbosa, 1998). Many conservation biocontrol tactics aim to increase densities of generalist predators (Koss et al., 2005; Symondson et al., 2002), which would be beneficial in alfalfa biocontrol because specialist predators are less common, and perhaps ineffective, in this crop. For example, in alfalfa a specialist parasitoid (Parastenus spp.) of Lygus hesperus was investigated for its control of Lygus, it was found to be less than promising (Waters et al., 2003). In Washington State parasitism was less than 10%, a level insufficient to keep Lygus below economic thresholds (Waters et al., 2003). For this reason generalist predators will likely be the main contributors to conservation biocontrol in alfalfa seed.

Generalist Natural Enemies

Studies have shown that generalist predators can be inferior to specialists because intraguild predation is particularly common among generalists (Polis et al., 1989; Rosenheim, 1998; Snyder and Ives, 2001; Snyder and Wise, 1999), and because generalists possess a functional response that plateaus relatively rapidly (Sabelis, 1992). Also, generalists produce fewer offspring per prey consumed (Sabelis, 1992) and can be distracted by alternative, non-pest prey, from feeding on particular target pests (Hassell and May, 1986). Despite these limitations, generalist predators have been shown to successfully control pest populations in a majority of studies (Symondson et al., 2002).

However, the polyphagous behavior of generalist predators can also be desirable for biological control in some situations, because generalists can subsist on alternative prey when target pest populations are low (Ehler and Miller, 1978; Settle, et al., 1996). The first theoretical model involving alternative prey was published by Williamson (1957) and since then there has been a growing body of research both supporting and opposing alternative prey as an aid to biological control (Abrams and Matsuda, 1996; Chaneton and Bonsall, 2000; Harmon and Andow, 2004; Hazzard and Ferro, 1991; Holt and Lawton, 1994; Koss et al., 2004; Östman and Ives, 2003; Östman, 2004; Settle et al., 1996).

Alternative prey has been shown to have both positive and negative effects on biological control. In tropical rice, detritus-feeding and plankton-feeding insects have been demonstrated to sustain early season populations of generalist predators, giving the predators a "head start" on the later-developing target pest (brown rice leafhopper) (Settle, et al., 1996). However, it has also been demonstrated that alternative prey can disrupt biocontrol of target pests if generalist predators prefer the non-targeted prey, or exhibit switching behavior due to higher densities of alternate prey (Chesson, 1989; Symondson et al., 2002). In a study by Halaj and Wise (2002), detritus was added to a cucurbit cropping system in order to increase the abundance of alternative prey, which in turn might increase the abundance of natural enemies and improve the control of pests. However, cucurbit production was reduced even though the density of predators increased. Lower cucurbit production was thought to have been reduced by predators feeding on the more abundant detritivores, instead of cucurbit pests. In alfalfa seed production aphids are a common and abundant alternative prey, which might contribute or detract from Lygus biocontrol for the reasons described above.

Research Objectives

Our first objective in this study was to 1) document the predator fauna present in alfalfa fields in Washington State, 2) identify the key predators of Lygus, and 3) examine the role of aphid alternate prey on Lygus biocontrol. This research project contributes to the broader goal of reducing reliance on broad-spectrum insecticides in alfalfa seed production.

Addressed in this thesis are the following questions:

- How do natural enemy guilds differ in alfalfa fields treated with broadspectrum insecticides, versus fields treated with little or no insecticide?
- Which predatory insects feed on Lygus bugs?
- How is Lygus consumption affected by the presence of alternative prey?

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CHAPTER 2

COMPARISON OF INSECT GUILDS IN ALFALFA GROWN FOR SEED, VERSUS ALFALFA GROWN FOR HAY

INTRODUCTION

In Washington State the target pest for alfalfa seed production is the Lygus bug (Lygus spp.), and to a lesser extent alfalfa weevil Hypera postica (Gyllenhal), pea aphids Acyrthosiphon pisum (Harris), and other aphids (e.g. spotted alfalfa aphid Therioaphis maculata (Buckton) (Berg and Laurdale, 1982). Because Lygus bugs feed on seed tissue, growers have low tolerance for damage since this can reduce seed yields (Berg and Laurdale, 1982). Most growers rely on broad-spectrum insecticides for Lygus control (Berg and Laurdale, 1982). Since the inception of the Food Quality Protection Act (FOPA) the use of many insecticides, particularly broad-spectrum insecticides, have been, or will be, limited and/or relinquished (Epstein et al., 2000). The loss of insecticides is one of many reasons that alternative insect control methods are needed in alfalfa seed production. In addition, heavy reliance on insecticides can be detrimental because of secondary pest outbreaks and the development among pests of resistance to insecticides. For example, regular insecticide treatments can cause outbreaks of phytophagous mites in alfalfa (Cone, 1963). Further, previous studies have demonstrated that Lygus can quickly become resistant to many varieties of insecticides even within the same growing season (Snodgrass, 1996; Snodgrass and Scott, 2000). An additional problem with heavy reliance on insecticides is their negative impact on natural enemy populations (Croft, 1990; Hardin et al., 1995; Koss et al., 2005). Reducing the use of broad-spectrum insecticides can be instrumental in conserving arthropod natural enemies (Agnello et al., 2002). These natural enemies can then contribute to the biological control of insect pests.

Conservation biological control emphasizes the preservation of natural enemy populations. As defined by DeBach (1974), conservation of natural enemies involves manipulating the environment to retain natural enemies. This can be accomplished by removing or mitigating adverse factors, such as insecticides, or by providing resources, such as overwintering habitat (Hajek, 2004). Natural enemy conservation was the first form of biological control, dating back to 900 AD when Chinese citrus growers collected nests of predaceous ants to control foliar pests (Ehler, 1998). Since then, many other methods of predator conservation have been developed including habitat manipulation, the use of selective insecticides, and in the adoption of predator-friendly cultural practices (Barbosa, 1998; Landis et al., 2000). For example, a study in alfalfa hay revealed that leaving un-harvested strips of alfalfa retained natural enemies within the field, effectively conserving natural enemies (Hossain et al., 2000). Similarly, other studies have shown that the use of selective insecticides can conserve natural enemy populations (Koss et al., 2005; Agnello et al., 2002). Strip harvesting and selective pesticides are two methods for conservation biological control that could be implemented in alfalfa seed production in Washington State.

Our objective here was to investigate the potential for conservation biological control in alfalfa seed. In this study natural enemy guilds were assessed in both seed and hay fields. Hay fields have very low insecticide inputs (Chapter 1). Thus, by examining the predator guilds in both hay and seed alfalfa, we can speculate on the composition of the predator community in seed fields following in the adoption of reduced insecticide input as a predator conservation tactic.

MATERIALS AND METHODS

Study Sites

Study sites in 2003 and 2004 were located in central and south central Washington State. In 2003 there were 7 alfalfa seed fields and 3 alfalfa hay fields sampled; all 10 sites were located in Walla Walla County and centered around the city of Touchet. In 2004 there were 6 alfalfa seed fields and 4 alfalfa hay fields; these field sites were split between two counties, Walla Walla and Grant (Table 2.1). Fields in Grant County were centered around the city of Warden. Differences between the Touchet and Warden fields included different irrigation methods and crop timing in relation to weather conditions. Touchet, located in the southern part of the state, experiences warmer temperatures earlier in, and higher average temperatures throughout, the growing season. In Touchet, alfalfa seed fields were sprinkler irrigated only at the beginning of the year, while hay fields were irrigated throughout the season. In the Warden area, alfalfa seed fields were irrigated regularly until bees begun to forage; both center-pivot irrigation and ground irrigation methods were used in different fields. Alfalfa hay was harvested roughly every 3 to 6 weeks, for a total of at least 3 cuttings. Alfalfa seed was harvested once at the end of the growing season in August, in both seed-growing regions.

Foliar Invertebrate Sampling

Foliar invertebrates were sampled using a D-vac suction sampler (D-vac Company, Ventura, CA 93002, USA). All fields were D-vac sampled three times during the growing season, in May, June, and July. There were 10 D-vac samples per field, per sampling date. The field was entered in a random location and a zigzagging path, punctuated by the collection of samples, was followed toward the center of the field. Each D-vac sample consisted of 10 stops; at each stop the cone was placed over an area of foliage for 3 seconds, and shaken to dislodge insects clinging to the foliage. The distance between each stop was 10 meters. Collection bags were placed into coolers with ice packs for transport to the lab. Once at the lab, samples in D-vac bags were transferred into 1-gallon Ziplock brand bags and stored in a freezer, until they could be sorted, identified and counted at a later date.

Ground Invertebrate Sampling

The ground fauna was sampled using pitfall traps. As with Dvac sampling, all fields were sampled using pitfall traps three times, once each in May, June and July. Pitfall traps were constructed using 12 oz. Dixie brand plastic cups, 8 in. plastic disposable plates and a piece of 8-cm long, 16-gauge wire . Holes were dug and cups were inserted flush into the ground, and plastic plates were held above the cup with the piece of wire in order to prevent debris and water from entering the trap. In 2003 there were 40 pitfall traps per field, and anti-freeze was used as a preserving agent. In 2004 there were 10 traps per field and samples were collected live. Pitfall traps were 10 meters apart and placed haphazardly between plants. Traps were left in fields for varying

amounts of time (Table 2.2). When traps were emptied the collected arthropods were placed in 1-gal Ziplock bags and transported to the lab in a cooler where the material was identified and counted. When traps were not in use they were covered by an inverted cup. Because of wide variation in pitfall trap catch intervals, and in the number of recoverable (undamaged) traps during each sampling interval, catch per day per recovered trap was calculated by averaging the total catch by the number of traps recovered, and the number of days those traps were open. Inconsistencies in the number of recovered traps were largely the result of trap loss due to flooding and/or damage by tractors or other farm equipment.

Statistics

Data for D-vac and pitfall samples were analyzed using repeated measures MANOVA in SYSTAT (\$PSS, Chicago, Illinois, USA). Where time x year x type interaction was significant, we conducted individual ANOVAs for each sampling date. D-vac data for Lygus, pea aphid and foliar predators were analyzed separately.

RESULTS

Lygus

The field type x time interaction was marginally significant (Wilks' ? = 0.674, $F_{2,15} = 3.629$, p = 0.052), because Lygus populations increased generally increased more dramatically in hay than seed fields. Overall, Lygus densities were higher in hay than seed fields ($F_{1,16} = 18.32$, p = 0.001; Figure 2.1 a and b). There was a significant effect

for time (Wilks' ? = 0.531, $F_{2,15} = 6.617$, p < 0.01), because Lygus populations increased through the season (Figure 2.1 a and b). All other factors and interactions were not significant (Table 2.3).

Pea Aphid

Of the two main effects, only year was significant ($F_{1,16} = 5.181$, p = 0.037); pea aphid populations in 2003 were much higher than in 2004 (Figure 2.1 c and d). All other factors and interactions were not statistically significant (Table 2.3).

Foliar Predators

Due to a significant interaction of time x year x type (Figure 2.1c, Wilks' ?= 0.585, $F_{2,15} = 5.315$, p = 0.018), we performed a separate ANOVA for each sampling date. For the May sampling date neither year nor type had a significant effect on predator populations (year $F_{1,16} = 0.785$, p < 0.01; type $F_{1,16} = 1.62$, p = 0.02). However, in June 2003 predator populations were higher in hay fields than in seed fields, but this difference was not evident in 2004 (year $F_{1,16} = 6.61$, p = 0.02; type $F_{1,16} = 9.48$, p < 0.01; type x year type $F_{1,16} = 10.17$, p < 0.01). In July 2003 predator populations were higher than in July 2004 (year $F_{1,16} = 4.80$, p = 0.04). Predator populations were marginally higher in hay fields than seed during July of both years (type $F_{1,16} = 4.06$, p = 0.06; type x year $F_{1,16} = 0.852$, p = 0.37). The 6 most common predators were *Gecoris* spp., *Orius* spp., *Nabis* spp., *Crysopera* spp., Thomisid spiders and Coccinellid beetles (Table 2.4 and Figure 2.2).

Ground Predators

Ground predator densities in seed and hay were similar during 2004, increasing over the course of the season. In 2003 predator densities were higher in hay during May and June but lower than seed in July. These differences among fields in the two sampling years accounted for the significant year effect ($F_{1,15} = 11.957$, p = 0.004) and time x year interaction (Wilks' ? = 0.531, $F_{2,14} = 6.193$, p = 0.012). The 6 most common ground predators were centipedes, Linyphild spiders, *Bembidion* spp., medium sized Staphylinid beetles, *Calosoma* spp. and *Amara* spp. (Table 2.4 and Figure 2.4).

DISCUSION

From survey data collected in 2003 and 2004 from alfalfa seed and hay fields, Lygus, pea aphid and predators densities were analyzed. D-vac data revealed that Lygus populations tend to increase through the growing season. Lygus populations were higher in hay fields than in seed fields, and were higher in 2003 than in 2004. Pea aphid populations in hay and seed fields did not differ. As with Lygus, pea aphid populations were higher in 2003 than in 2004. Predator densities in D-vac samples differed by field type, year, and time. Foliar predator densities in seed fields remained fairly stable, while densities in hay fields varied from May to July. Ground-dwelling predator populations in contrast tended to increase through the growing season in fields of both types, except for hay fields in 2003. Ground predator densities were higher in 2004 than in 2003 and did not differ between hay or seed fields.

Alfalfa seed growers are concerned about Lygus populations throughout the year; however, there is an increased emphasis on control once seed formation begins. Most seed pods are formed in July, which is when Lygus populations tend to peak. During July the most common predators are *Hippodamia convergens, Coccinella septempunctata, Calosoma* spp., Staphylinid beetles, *Nabis* spp., Thomisid spiders and Linyphiid spiders. While Lygus populations were higher in hay fields in 2003, in 2004 there was no difference between the two types of fields. In addition, average Lygus densities in hay during 2004 were half the average population density in 2003. The higher predator densities in hay in July 2004 could be underlying these trends, suggesting that generalist predators may be suppressing Lygus populations in hay. These results are promising for conservation biological control in seed.

Several studies comparing high input and low, or no, input insecticide regimes have found that predator densities are highest in low or no input fields (Agnello et al., 2002; Hilbeck and Kennedy, 1996; Koss et al., 2005; Letourneau and Goldstein, 2001). Koss et al. (2005) performed a study, in Washington potatoes, where management regimes were compared between conventional fields that used broad-spectrum insecticides, soft fields that used selective insecticides, and organic fields that used certified organic insecticides. Results from this study indicated that organic fields had the highest predator densities, but also had the highest densities of pests. Fields treated with selective pesticides also had high predator densities, similar to those seen in organic fields, but in fields treated with selective insecticides pest densities were similar to those seen in conventionally-managed fields. Fields treated with selective insecticides also maintained had high yields, similar to those seen in conventional fields. Thus, the study

of Koss et al. (2005) supports the use of selective insecticides, because fields treated with selective insecticides combined high predator densities, low pest densities, and high yields. A 3 year study in apple orchards by Agnello et al. (2002) compared the use of selective versus broad-spectrum pesticides, for the control of aphid and mite pests. The study documented that reduced use of pesticides, coupled with using less toxic pesticides, conserved natural enemies. By the end of the third year of their study, orchards treated with selective pesticides had harvests that were equal or superior to those in conventionally treated orchards (Agnello et al., 2002). Based on these previous studies, and the results of our survey of seed and hay alfalfa fields, seed growers may be able to conserve predators and control Lygus by using selective insecticides, once selective insecticides targeting Lygus are developed. However, since most of the predators collected in our survey were generalists, additional studies will be required to identify which of the predator species in this diverse community actually feed on Lygus.

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Year	Name	Field	Туре	Irrigation	Duration of
		Location			Irrigation
2003	7 th Day North	Touchet	Seed	standard	Early season
	7 th Day South	Touchet	Seed	standard	Early season
	7 th Day Hay	Touchet	Hay	standard	Throughout
	Moore North	Touchet	Seed	standard	Early season
	Moore South	Touchet	Seed	standard	Early season
	Moore Hay	Touchet	Hay	standard	Throughout
	Lower Dry Creek	Touchet	Seed	standard	Early season
	Office	Touchet	Hay	standard	Throughout
	McDonald	Touchet	Seed	standard	Early season
	Byrnes	Touchet	Seed	standard	Early season
2004	Moore North	Touchet	Seed	standard	Early season
	Moore East	Touchet	Seed	standard	Early season
	Dewy	Touchet	Hay	standard	Throughout
	Field by Wheat	Touchet	Hay	standard	Throughout
	Providence	Warden	Seed	ground	Midseason
	Long	Warden	Seed	ground	Midseason
	Rd 6	Warden	Seed	center-pivot	Midseason
	Rd X	Warden	Seed	center-pivot	Midseason
	Rd 3	Warden	Hay	center-pivot	Throughout
	Rd 6 hay	Warden	Hay	center-pivot	Throughout

Table 2.1. Alfalfa field sites by year, location, type and irrigation.

Date	# of traps per field	# sorted	Days out
May 2003	40	20	10-15
June 2003	40	20	10-15
July 2003	40	20	10-15
May 2004	10	10	10
June 2004	10	10	10
July 2004	10	10	10

Table 2.2 – Logistics of pitfall samples.

	Lygus	Pea aphid	Foliar Predators	Ground Predators
type	F _{1,16} =18.32 p=0.001	F _{1,16} =0.060 p=0.810	F _{1,16} =12.030 p=0.003	F _{1,15} =0.150 p=0.704
year	F _{1,16} =5.442 p=0.033	F _{1,16} =5.181 p=0.037	F _{1,16} =6.796 p=0.019	F _{1,15} =11.957 p=0.004
type x year	F _{1,16} =7.450 p=0.015	F _{1,16} =0.007 p=0.934	F _{1,16} =5.785 p=0.029	F _{1,15} =0.144 p=0.710
time	Wilks' ?=0.531	Wilks' ?=0.912	Wilks' ?=0.631	Wilks' ?=0.430
	F _{2,15} =6.617 P=0.009	F _{2,15} =0.726 P=0.500	F _{2,15} =4.378 P=0.032	F _{2,14} =9.267 P=0.003
time x type	Wilks' ?=0.674	Wilks' ?=0.890	Wilks' ?=0.726	Wilks' ?=0.998
	F _{2,15} =3.629 P=0.052	F _{2,15} =0.929 P=0.417	F _{2,15} =2.831 P=0.091	F _{2,14} =0.012 P=0.988
time x year	Wilks' ?=0.952	Wilks' ?=0.849	Wilks' ?=0.613	Wilks' ?=0.531
	F _{2,15} =0.382 P=0.689	F _{2,15} =1.333 P=0.293	F _{2,15} =4.738 P=0.025	F _{2,14} =6.193 P=0.012
time x year x	Wilks' ?=0.713	Wilks' ?=0.854	Wilks' ?=0.585	Wilks' ?=0.929
type	F _{2,15} =3.016 P=0.079	F _{2,15} =1.280 P=0.307	F _{2,15} =5.315 P=0.018	F _{2,14} =0.532 P=0.599

Table 2.3 – Statistical analysis for D-vac and pitfall samples.

Table 2.4 – Insects identified Itom D-vac and pittan samples in 20		D:#f=11
Insects identified	D-vac	Pitfall
Collembola	present	present
Ephemeroptera	present	0
Orthoptera		
Gryllidae (cricket)	1	13
Gryllotalpidae (mole cricket)	0	present
Stenoplematidae (Jerusalem cricket)	0	present
Tettigoniidae (katydid & Mormon cricket)	2	0
Acrididae (short-horned grasshopper)	3	0
Mantodea		
Mantidae (mantid)	present	0
Dermaptera		
Forficulidae		
Forficula auricularia (European earwig)	0	166
Thysanoptera		
Ăeolothripidae		
Aeolothrips fasciatus Banded-wing thrips	present	0
Thripidae		
Frankliniella occidentalis (Pergrande) western flower thrips	present	present
Scolothrips sexmaculatus (Pergande) sixspotted thrips	present	0
Hemiptera		
Anthocoridae		
Orius spp.	309	present
Lygaeidae		procent
Geocoris spp.	1448	present
Miridae		procent
Lygus spp.	1436	present
Other	8	present
Nabidae		procont
Nabis spp.	177	present
Pentatomidae		present
Acrosternum hilare (green stink bug)	1	0
Reduviidae (assassin bug)	4	0
Cicadellidae	<u>т</u>	0
Circulifer spp. and Empoasca spp.	3792	present
Aphididae	J172	present
Apriluluae Acyrthosiphon kondoi Shinji (blue alfalfa aphid)	present	0
Acyrthosiphon pisum (Harris) pea aphid	39010	present
Acymosphon pisum (nams) pea aphid Aphis craccivora Koch (cowpea aphid)	111 (2003)	0
		Ű
Aphis fabae Scopoli (bean aphid)	present	0
Therioaphis maculate (Buckton) spotted alfalfa aphid	831(2003)	0
Neuoptera		
Chrysopidae		05
Chrysopa carnea Stephens (common green lacewing)	119	35

Table 2.4 – Insects identified from D-vac and pitfall samples in 2003 and 2004.

Hemerobiidae		
Sympherobius barberi (Banks) barber brown lacewing	present	0
Mymeleontidae		
Dendroleon obsoletus (Say) spottedwinged antlion	present	1
Lepidoptera	12	32
Peiridae		
Colias eurytheme Boisduval (alfalfa caterpillar)	present	present
Noctuidae		procent
Autographa californica (Speyer) alfalfa looper	present	present
Pyralidae	p.000111	p
Loxostege cereralis (Ziller) alfalfa webworm	present	present
Coleoptera	p.000111	p
Predatory larva	0	24
Anthicidae	Present	present
Carabidae		
Amara spp.	0	119
Bembidion spp.	0	374
Calathus spp.	0	104
Calosoma spp.	0	124
Pterostichus spp.	0	72
Other	0	55
Coccinellidae		
Coccinella septempunctata Linnaeus (sevenspotted lady beetle)	54	present
Hippodamia convergence	20	present
Coccinella transversoguttata richardsoni Brown (transverse lady	4	
beetle)		
Coccinella septempunctata and Hippodamia convergence*		29
Curculionidae		
Hypera brunnipennis (Boheman) Egyptian alfalfa weevil	present	present
Hypera postica (Gyllenhal) alfalfa weevil	present	9
Hypera punctata Fabricius (clover leaf weevil)	present	present
Otiorhynchus ligustici (Linnaeus) alfalfa snout beetle	present	330
Sitona hispidulus (Fabricius) clover root curculio	present	729
Other	0	3
Elateridae (click beetle)	present	0
Meloidae (blister beetle)	present	0
Scarabidae (dung beetle)	0	2
Silphidae	0	22
Staphylinidae (rove beetle)		
micro	present	52
small	present	93
medium	present	168
larg	present	45
Tenebrionidae	present	0
Coleoptera Other	28	

Diptera		
Syrphidae	present	present
Tachinidae	present	present
Hymenoptera		
Braconidae		
Aphidius spp.	present	0
Praon spp.	present	0
Eurytomidae		
Bruchophophagus roddi (Gussakovsky) alfalfa seed chalcid	present	present
Formicidae	present	1400
Diplopoda (millipedes)	0	13
Chilopoda (centipedes)	0	1394
Arachnida		
Araneidae (orb weavers)	16	5
Linyphiidae (dwarf spiders)	38	874
Lycosoidae (wolf spiders)	0	94
Thomisidae (crab spiders)	86	19
Hunting other	0	156

Present indicates a visual identification of the species in the field or that it was collected but not counted.

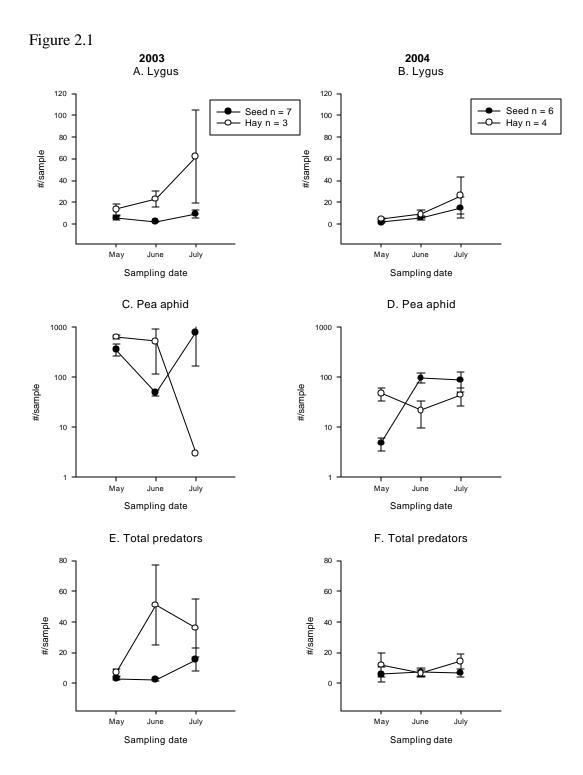
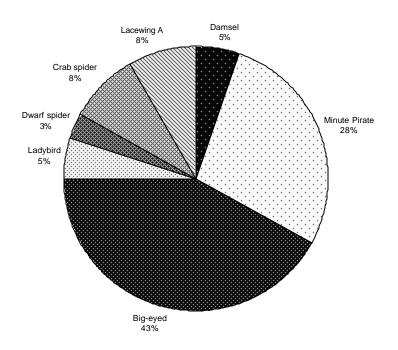
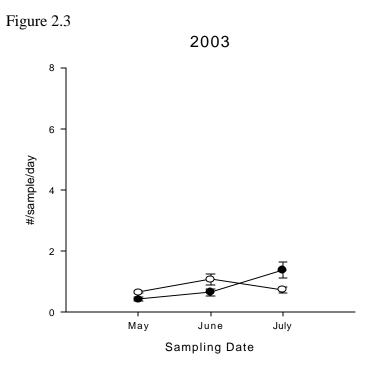


Figure 2.2





● Seed ● Hay



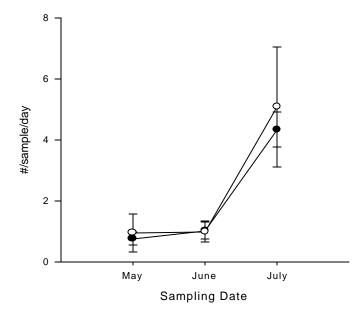
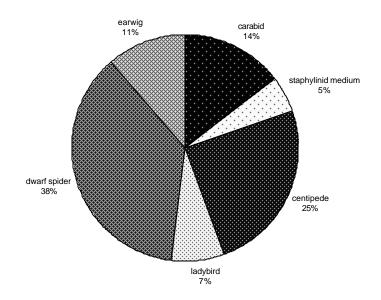


Figure 2.4



CHAPTER 3

PEA APHIDS MAINTAIN LEVELS OF GENERALIST PREDATORS FOR CONTROLLING LYGUS PESTS IN ALFALFA GROWN FOR SEED

INTRODUCTION

Throughout the history of biological control of insect pests, there have been differing opinions on the efficacy of generalist predators as control agents. Studies have shown that generalist predators can be inferior to specialists because of generalists' frequent participation in intraguild predation (Polis et al., 1989; Rosenheim, 1998; Snyder and Ives, 2001; Snyder and Wise, 1999), a functional response that rapidly plateaus (van den Meiracker, and Sebelis, 1992), fewer offspring per prey consumed (Sabelis, 1992), and generalists' tendency to feed on alternative, non-pest prey rather than pests (Hassell and May, 1986). However, despite these limitations, generalists have been shown to successfully control pest populations in most empirical studies: in a recent review by Symondson, Sunderland and Greenstone (2002), generalist predators were found to be successful in significantly reducing the number of pest species in about 75% of manipulative field experiments.

The polyphagous behavior of generalist predators is sometimes a desirable character for biological control, due to generalists' ability to subsist on alternative prey when focal prey densities are low (Ehler and Miller, 1978; Settle, et al., 1996). When prey densities are low, generalist predators adopt a lying-in-wait strategy (Den Boer,

38

1982), so that when pests rebound they can aggregate to the pest (Bryan and Wratten, 1984). The first theoretical model involving alternative prey was published by Williamson (1957); ever since, there has been a growing body of research presenting scenarios where alternative prey both contribute and detract from biological control of a target pest (Abrams and Matsuda, 1996; Chaneton and Bonsall, 2000; Harmon and Andow, 2004; Hazzard and Ferro, 1991; Holt and Lawton, 1994; Koss et al., 2004; Östman and Ives, 2003; Östman, 2004; Settle et al., 1996).

A classic example of how alternate prey can be beneficial to biocontrol comes from the work of Settle et al. (1996), working in a rice agroecosystem. In tropical rice, detritus- and plankton-feeding insects have been demonstrated to sustain early season populations of generalist predators, giving the predators a "head start" on the laterdeveloping targeted pest, brown rice planthopper (Nilaparvata lugens Stal) (Settle et al., 1996). The brown rice leafhopper is an example of a negative prey-prey interaction, where the targeted pest is being negatively affected by the presence of the alternative prey through augmented predator densities and thus increased predation on the target (Figure 2.1). However, it has also been demonstrated that alternative prey can disrupt biocontrol of target pests, if generalist predators prefer the non-target prey, or exhibit switching behavior (from the target to the alternative prey) (Chesson, 1989; Symondson, et. al, 2002). When a positive predator-predator interaction of this type occurs, biological control of the targeted pest is weakened (Figure 2.1). For example, Halaj and Wise (2002) added detritus to a cucurbit cropping system, in order to increase the abundance of detritivore alternative prey. It was hoped that augmented detritivores would provide additional prey for predators, raising predator densities and increasing attacks on the

target herbivore. However, although the density of predators did indeed increase when detritivore densities were higher, pest densities remained high and cucurbit production was not improved. Apparently, predators preferred feeding on the more abundant detritivores, rather than the targeted cucurbit pests.

In alfalfa there is a rich fauna of insects, both predators and pests (Pimentel and Wheeler, 1973). The key pest of alfalfa grown in Washington State is the Lygus bug (Hemiptera: Miridae) (Berg and Lauderdale, 1982). Lygus adults can become problematic early in the season because they overwinter in plant debris left in alfalfa fields and can achieve large populations early in the season (Schaber and Entz, 1994). Another source of Lygus is from adjacent hay fields; when hay is harvested Lygus often emigrate to seed fields (Schaber et al., 1990). Also, in addition to feeding on plant phloem, Lygus feed on reproductive tissue, damaging seeds (Gupta, 1979; Gupta et al., 1980). Lygus feeding thus results in seed abortion, plant stunting and seed shrivel (Gupta, 1979; Gupta et al., 1980; Shull et al., 1934; Sorenson, 1936). Due to increased pesticide resistance in Lygus populations (Crafton-Cardwell et al., 2000), as well as reduced availability of insecticides (Scheuplein, 1999), there is need for alternative methods to control Lygus. Biological control is a proven alternative method (Debach, 1974; Chang and Kareiva, 1999; Hajek, 2004; Van Den Bosch, 1971) that perhaps could be applied to alfalfa seed production.

In addition to Lygus, aphids are also a commonly occurring herbivore in alfalfa seed fields, and although several different aphid species attack alfalfa locally, pea aphids (*Acrythosiphon pisum* Harris) are the most common species (Gupta, 1979). Pea aphids primarily feed on the stem or leaf of the plant, rather than the growing tips or seeds, making aphids less damaging than are Lygus (Blodgett, 2003; Muegge, 2003); moderate aphid populations can be present on the plant without causing economically-significant damage (Rincker et al., 1987). Pea aphids can often be more abundant than are Lygus (See Chapter 2), and aphids frequently exhibit population outbreaks associated with abiotic factors (e.g., weather and insecticide applications) (Blodgett, 2003); during these outbreaks pea aphids may act as alternative prey for generalist predators (Blodgett, 2003).

The goal of our study was to evaluate the potential for aphid alternative prey to disrupt control of Lygus, our focal pest. A previous predator-pest survey of alfalfa fields (Chapter 2) revealed that the most abundant predators during peak Lygus densities were *Hippodamia convergens, Coccinella septempunctata, Calosoma* spp., Staphylinid beetles, *Nabis* spp., Crab spiders (*Misumenops* spp.), and Linyphiid spiders. We conducted a series of feeding assays to determine the interactions among these predators, Lygus, and pea aphids. First, an initial no-choice assay of the common predators was conducted to identify which species ate the largest numbers of Lygus within a timed interval. Second, a microcosm assay was run to determine how Lygus predation by the two most effective predators, nabid bugs and crab spiders, would be affected by the presence of aphids.

METHODS AND MATERIALS

Collection of Study Organisms

Study organisms were collected from production alfalfa fields in south-central and central Washington State. Ground-dwelling insects were collected using pitfall traps (described in Chapter 2). Ten to twenty pitfall traps were set up in a field 2 days before the beginning of the experiment, and left open overnight. Foliar insects were collected using a D-vac suction sampler, 24-h prior to the experiment, from the same fields where pitfall collections were made. All live insects were transported in a cooler containing icepacks, to the lab, where they were identified and sorted into individual vials and stored in a dark incubator at 8° C until use in experiments, ca. 24-h later.

No-choice Petri Dish Experiment

No choice-feeding assays were conducted to identify the most effective Lygus predators. Assays were conducted in 100 x 15 mm plastic Petri dishes containing a 4-cm piece of washed (organic) green bean Then, either a single 2^{nd} to 3^{rd} instar pea aphid, or a 2^{nd} to 3^{rd} instar Lygus nymph, was placed into each arena. The green bean was essential to keep Lygus nymphs alive (A. E. Jorgensen, unpublished data), and for consistency pea aphid arenas also received a piece of bean. An individual predator was added to each Petri dish. Petri dishes were stored in an incubator at $21 \pm 2^{\circ}$ C, 16:8 photoperiod. Arenas were observed every 2 hours for 10 hours, for a total of 5 observations. During each observation if a pea aphid or Lygus was consumed a replacement was made; thus the maximum number of prey that could have been consumed during this assay was 5. There were between 4 and 12 replicates, based on the number of predators of each taxa collected within the day preceding each feeding trial, for each combination of predators and Lygus or pea aphid.

Microcosm Experiment

The effect of aphids on Lygus predation by the two most effective Lygus predators was assessed in larger arenas. Here, microcosms consisted of a 32 oz. clear plastic deli container, with a 3/8 in hole drilled in the bottom of each container, into which a 9-DRAM scintillation vile was screwed. The lid of the container was punctured with 16 holes. Twelve hours prior to the start of the experiment, a fresh alfalfa stem was added to each microcosm. Stems were obtained from an unsprayed alfalfa field on the Pullman campus, trimmed to 10 cm and with leaves thinned to three per stem. Stems were then wrapped with 3 sheets of tissue paper and inserted into the scintillation vial at the base of the container. Water was added to the vial to moisten the tissue. A 2-cm deep layer of sterilized sand was added to the bottom of the deli container in order to cover the top of the vial, and provide a more natural foraging surface for the predators. On the morning of the experiment, Lygus and pea aphids were introduced into each arena according to treatment, and allowed to acclimate for 2 h. There were six treatments: 1) Lygus only control, 2) Lygus + aphid control, 3) Nabis + Lygus, 4) Nabis + Lygus + aphid, 5) Crab spider + Lygus, and 6) crab spider + Lygus + aphid. Thus, the experiment encompassed a complete 3 x 2 factorial design. The experiment was run for 24 hours, at 16:8 L:D and $24 \pm 2.7^{\circ}$ C, on greenhouse benches. At the end of the experiment the microcosms were destructively sampled for remaining insects, with the bottom and sides of the microcosm visually searched for any insects not on the foliage. The experiment was run between June and July of 2004, and there were 3 blocks each including ten replicates for each treatment.

Statistics

Data for each prey in the no-choice assay were analyzed separately using one-way ANOVA, followed by Tukey-Kramer HSD post-hoc tests (a = 0.05). Microcosm data were analyzed in a 3 factor (block, predator and alternative prey) ANOVA model. All data were analyzed in SYSTAT (SPSS, Chicago, Illinois, USA).

RESULTS

No-choice Petri Dish Experiment

The objective of this experiment was to determine which of the 6 commonly occurring generalist predators ate Lygus and pea aphids, and in what quantities. Predators differed in Lygus consumption rate ($F_{1,5} = 3.518$, P = 0.009; Fig. 2a); *Nabis* spp. ate the most Lygus (Fig. 2a), while linyphiid spiders ate the fewest (Fig. 2a). Predators also significantly differed in their feeding rates on pea aphids ($F_{1,5} = 12.037$, P < 0.001); *C. septempunctata* consumed the most pea aphids (Fig. 2b), while crab spiders consumed the fewest pea aphids (Fib. 2b). The predator that consumed the second highest average of Lygus was the crab spider (Fig. 2b). Since *Nabis* and crab spiders consumed the largest number of Lygus, they were chosen for use in the microcosm experiment to compare the affect of alternative prey on their consumption of Lygus.

Microcosm Experiment

This experiment was designed to determine if alternative prey would affect the consumption of Lygus by *Nabis* and crab spider predators. There was a significant predator effect ($F_{2,162} = 47.316$, p < 0.001; Table 2.1; Fig. 3); fewer Lygus were consumed in predator treatments than in the control (Tukey-Krammer HSD; p < 0.05; Fig. 3), but there was no difference in Lygus consumption between crab spider and nabid predators (Tuckey-Krammer HSD; p > 0.05; Fig. 3). When alternative prey were present, significantly more Lygus survived ($F_{1,162} = 10.263$, p = 0.002; Table 2.1; Figure 2.2). There was a significant block effect ($F_{2,162} = 5.497$, p = 0.005; Figure 2.2; Table 2.1), with fewer Lygus surviving in Block 2.

DISCUSSION

In our alfalfa system the targeted pest, Lygus, and alternative prey, pea aphid, are present in high numbers at the same time (Chapter 2). We initially thought that pea aphids might disrupt biocontrol of Lygus by generalist predators, because of the temporal overlap in the two prey taxa. In many studies alternative prey have been reported to interfere with biological control (Chesson, 1989; Hazzard and Ferro, 1991; Abrams and Matsuda, 1996). For example, in a study regarding the biological control of a mosquito pest, *Culex pipiens*, by a notonectid bug, *Notonecta hoffmani*, the presence of non-target Drosophila flies reduced consumption of the target pest (Chesson, 1989). In our study, of the generalist predators that are present during peak Lygus populations, *Nabis* and crab spider predators consumed the greatest numbers of Lygus over a 24 hour feeding period.

Our follow-up microcosm experiment revealed that Lygus survivorship increased in the presence of aphids. This effect appeared to be driven by higher survivorship of Lygus in all treatments including aphids, regardless of predator presence. It is unclear why aphids generally improved Lygus survivorship; regardless, based on these results we would predict higher Lygus survival in fields containing robust aphid populations However, it remains possible that at a longer time scale, predators will be attracted to pea aphids increasing Lygus biocontrol thru positive prey-prey interactions (Östman and Ives, 2003).

Our results are inconsistent with other studies in agroecosystems that demonstrate a benefit of alternative prey on biocontrol of pests even when alternative prey and pests co-occur. A study by Harmon et al. (2000) investigated predation of pea aphids by the omnivore *Coleomegilla maculata* with varying densities of dandelions. It was shown that increased dandelion densities were associated with higher densities of *C. maculata* and lower densities of pea aphid. The study demonstrated that a pollen-producing plant indirectly decreased herbivore densities by attracting an omnivorous predator – once moving into a patch to feed on dandelion pollen, the predators also £d on aphids. The quality of alternative prey versus focal prey is important when assessing indirect interactions. In a study by Eubanks et al. (2000), the predator *Geocoris punctipes* was presented with an immobilized aphid and a mobile aphid, the predator preferred to feed on the mobile aphid. The predator still preferred to feed on the pea aphid despite its lesser quality.

One possibility not explored in this study is the increased chance of emergent impacts in the multiple predator-aphid-Lygus web. Emergent impacts, such as synergism,

can result in greater levels of pest control in combination with multiple predators. In a classic example, Losey and Denno (1998) examined carabid and coccinellid beetles for efficacy of reducing pea aphid populations in alfalfa. Carabids alone had no effect on reducing pea aphid population; however, ladybird populations were capable of reducing pea aphid populations. However, when the two predators were used together to control aphids there was a synergistic interaction between the predators. In the presence of foraging ladybird beetles pea aphids exhibited an anti-predator defense, falling to the ground to escape the ladybirds. But, once on the ground pea aphids became vulnerable to attack by carabid beetles. Thus, the two predators together were more effective together than would be predicted from simply adding their individual impacts. Multiple predators could have similar effects on Lygus in the field, where Lygus will be attacked by a diverse community of predators. When Lygus nymphs are disturbed they scurry (Cranmer, 2004); this increases their potential encounter rate with predators, such as *Nabis* and crab spiders. Together, actively foraging predators and ambush predators could be acting synergistically to control Lygus.

<u>Summary</u>

Grower tolerance of moderate pea aphid populations may enhance Lygus control by some predators (e. g. *Nabis*). Transition to selective pesticides will further conserve generalist predators, which may also feed on secondary pests. Through careful management, pea aphids might be used to enhance conservation biocontrol of Lygus.

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Nabis and Crab spider microcosm experiment

Treatment	Degrees of freedom	F	p-value
Blocks	2,162	5.497	0.005
Alt	1,162	10.263	0.002
Pred	2,162	47.316	< 0.001
Block*Alt	2,162	0.390	0.678
Block*Pred	4,162	0.691	0.599
Alt*Pred	2,162	1.416	0.246
Blocks*Alt*Pred	4,162	0.358	0.838

TABLE 3.1 – Statistics from analysis using ANOVA with 3 variables, Block, Alt, and Pred. Treatments: Block – block effect; Alt – presence or absence of alternative prey; Pred – control, nabid, or crab spider.

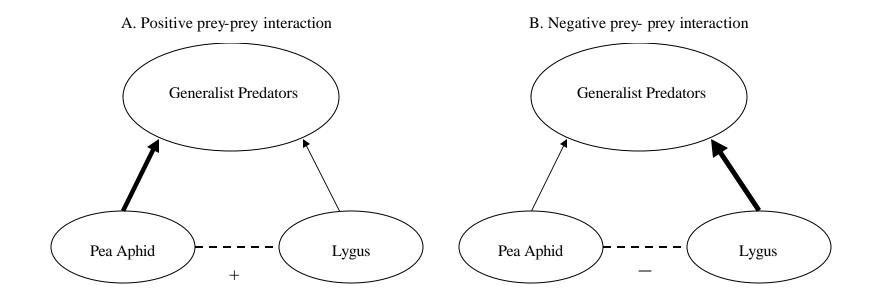


FIGURE 3.1 – There are two ways pea aphids can affect Lygus predation by generalist predation. (A) Presence of aphids can decrease predation on Lygus, a positive indirect prey-prey interaction. (B) Presence of aphid prey can increase the predation upon Lygus, a negative prey-prey interaction (apparent competition). Solid arrows indicate trophic or direct interactions, arrow pint in the direction of energy flow. Dashed lines indicate indirect effects.

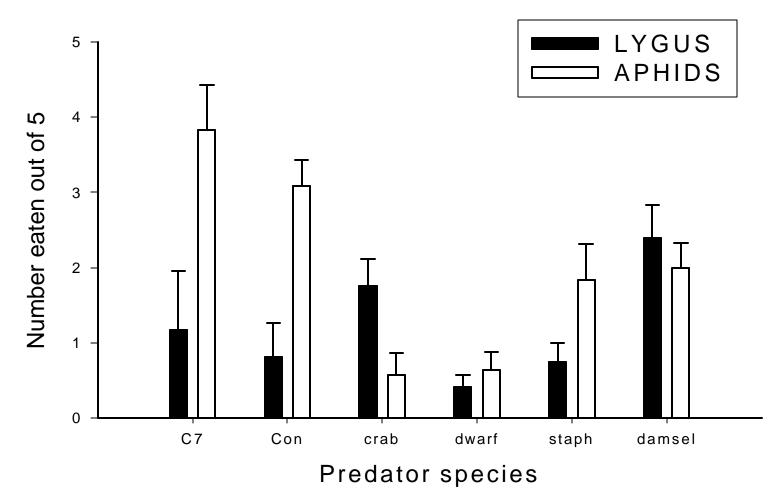


FIGURE 3.2 – Number of Lygus or pea aphids eaten out of 5 the six most commonly occurring generalist predators in alfalfa fields in Washington State.

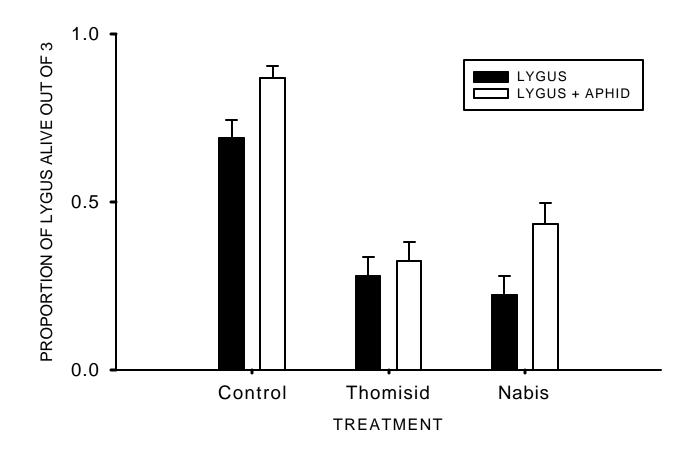


FIGURE 3.3 – Effect of aphid alternate prey on Lygus predation by the two generalist praetors commonly occurring in alfalfa fields. Crab spiders and the predatory bugs *Nabis* spp. Bars represent mean \pm s.e. proportion of Lygus eaten (out of 3) during 24-h. For each treatment n=30.