EFFECTS OF INSECTICIDE TREATED GREEN

PEACH (*M. PERSICAE*) APHIDS ON THE SURVIVAL

AND DEVELOPMENT OF COCCINELLIDAE

By

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Abstract: Annual canola yield losses have been significant in Oklahoma due to high insect pressure, and insecticide treatments are required multiple times throughout the growing season to produce a profitable crop. Synthetic pyrethroids are the most commonly used foliar insecticides for management of insect pests in canola, but they have been shown to have negative effect on both natural enemies and pollinators. Aphids in crop systems are a primary source of prey for lady beetles, making them beneficial in fields with frequent aphid outbreaks (Kring et al. 1985, Rice and Wilde 1988, Elliott et al. 1996, Jones 2001). This study will explore the effects of insecticide (flonicamid and sulfoxaflor)-treated aphids on ladybeetle development and survival.

Larval development time, adult body size, and male:female ratios from my trials indicate that flonicamid could be a valuable tool in fields where Coccinellids provide some level of biological control of aphids. Flonicamid allows for the possibility of longer-term lady beetle survivorship, where other pesticides such as synthetic pyrethroids do not. Conversely, these studies also indicates that sulfoxaflor treated aphids have a toxic effect on Coccinellids and may be a poor choice for use in similar fields. Consumption of sulfoxaflor treated aphids resulted in longer development times, which expose lady beetle larvae to other mortality effects, such as predation and further pesticide sprays. Sulfoxaflor treatments also resulted in reduced body size in adult lady beetles, which would likely decrease fecundity and ultimately reduce population size in the agricultural landscape. The fact that there were no surviving female *C. septempunctata* feeding on sulfoxaflor-treated aphids, and that male *H. convergens* were of smaller body sizes, indicates that sulfoxaflor-treated fields could be an ecological sink for these predator species.

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CHAPTER I

INTRODUCTION

Winter canola was introduced to Oklahoma as a rotational crop for winter wheat

in 2001. Canola in crop rotation allows for selective weed management and can improve

productivity in the agricultural system (Bushong et al. 2012). However, annual canola

yield losses have been significant in Oklahoma due to high insect pressure, and

insecticide treatments are required multiple times throughout the growing season to

produce a profitable crop. Synthetic pyrethroids are the most commonly used foliar

insecticides for management of insect pests in canola, but they have been shown to have

negative effect on both natural enemies and pollinators (Kaakeh, 1996; Jalali and

Leeuwen, 2009; Cloyd and Dickinson, 2006; Jansen et al., 2011). Canola might serve as a

sink for natural enemies in the landscape because of significant pyrethroid use and/or the

presence of unusable prey, and it is unclear if canola is appropriate habitat for predators

such as lady beetles. The most common group of insect pests in canola are aphids, and

natural enemies that feed on these aphids have been shown to exhibit decreased the

fitness, which can decrease the ability of those predators to provide significant biological

control.

Agrochemical companies have recently focused on the discovery and delivery of effective narrow-spectrum insecticides with unique modes of action that suppress insect pests while conserving beneficial organisms. Before these narrow-spectrum insecticides can be integrated into sustainable management programs, their environmental impacts must be documented on a landscape level. Flonicamid (Beleaf® FMC Corporation) and sulfoxaflor (Dow AgroSciences LLC) are two relatively new narrow-spectrum insecticides that have been recently registered for application in canola against plant-sucking insects. These insecticides are highly effective at suppressing Hemipteran pests, such as aphids, and should pose little to no risk for insect natural enemies and pollinators. Winter canola in the South Central US is an ideal crop to evaluate the environmental impacts of these unique compounds because aphid pests and natural enemies occur together annually in high numbers when insecticide use is justified.

The research reported in this thesis will describe how these new narrow-spectrum insecticides are compatible with current canola insect management programs. If beneficial insects can be maintained in canola fields, it may support populations in the agricultural landscape. Canola interfaces with wheat and pasture fields in Oklahoma and natural enemies such as lady beetles readily move between fields. The use of narrow-spectrum insecticides could allow for continued biological control services in these systems.

OBJECTIVES

The objectives of my thesis are:

1. Examine the effects of insecticide treated green peach aphids on the survival and development of Coccinellidae.

2. Investigate potential competitive interactions among Coccinellidae on insecticide treated canola plants.

CHAPTER II

LITERATURE REVEIW

Winter Canola

Since 2003, the land in the southern Great Plains devoted to growing winter

canola has increased to an estimated 300,000 acres in 2013 ("Crop Production"). Canola

is planted in September and October and flowers in early spring, providing floral

resources until seedpods form in late spring (Boyles and Sanders, 2003).

Canola seeds are harvested and crushed to produce oil and the remains are ground

into a protein supplement meal for livestock. Canola oil is approved by the FDA to

provide certain benefits to heart health (USCA, 2013), and has become the second most

used cooking oil in the United States. Its mild flavor, light consistency, and low saturated

fat have influenced its popularity and increased the demand on canola growers

worldwide.

Oklahoma is currently the second top producer of canola in the United States,

surpassed only by North Dakota and followed by Kansas, Colorado, Texas, and New

Mexico. The US Canola Association estimates that Southern Plains production will grow

to match the Northern Plains' 1.5 million acres by 2018 (USCA, 2014; Figure 1).
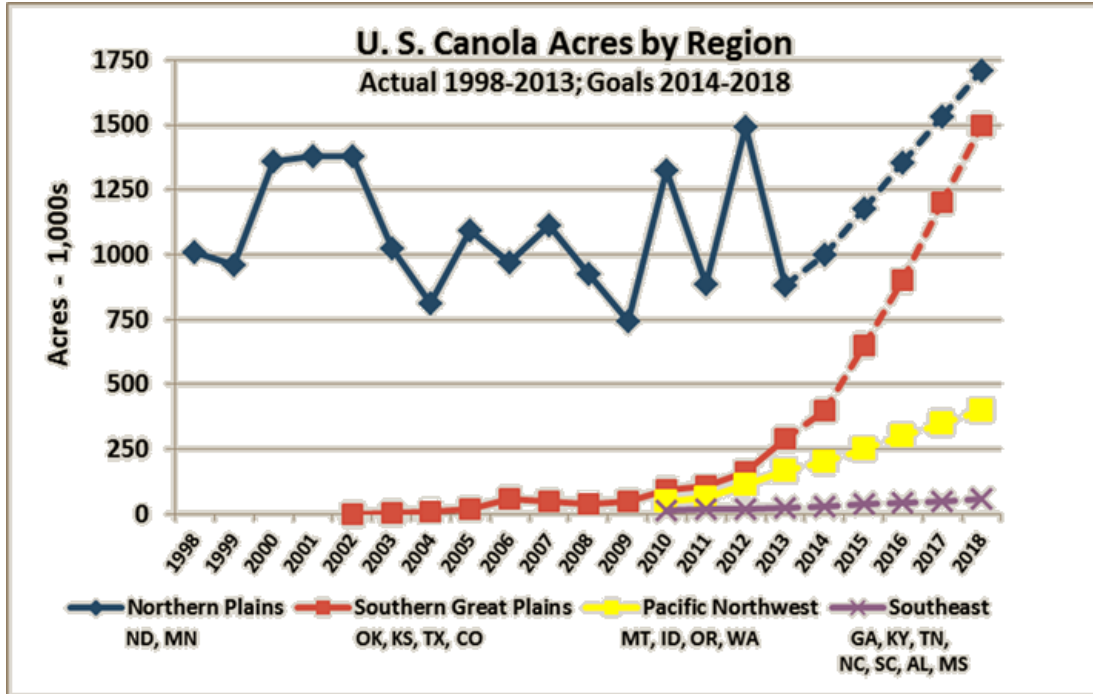
Figure 1. Actual acres of canola planted in the U.S from 1998-2013 and projected goals through 2018. [Courtesy of U.S. Canola Association]

Canola Pests

Aphid infestations in canola fields (Franke et al. 2009, Royer and Giles 2010) have resulted in low yields and complete crop loss (Boyles et al. 2004 and 2006) since canola was introduced into the southern Great Plains. Aphids reach damaging levels by early spring and causing significant losses to yield. Green peach (*Myzus persicae*), cabbage (*Brevicoryne brassicae*), and turnip aphids (*Lipaphis pseudobrassicae*) feed on the phloem of canola plants, causing wilting, yellowing of leaves, and decreasing the plant's vitality. Turnip aphids cause the greatest amount of damage, killing young canola plants from November until March, while Cabbage aphids damage during flowering and seedpod development (Boyles et al. 2006, Royer and Giles 2010). It is estimated that

each aphid on a canola plant can cause up to a half a pound of yield loss in the Great

Plains, and aphids generally exceed economic injury levels consistently every year

(Giles, unpublished observations).

Aphids have piercing-sucking mouthparts consisting of two sharp stylets and a

feeding tube. During feeding, the stylets and tube pierce through the plant epidermal cells

as the aphid secretes saliva into the puncture wound to begin digesting the cytoplasm and

sap. The aphid then exhibits probing behavior, piercing through the cell walls of the

plant's inner cell layer (palisade mesophyll cells) until it locates the vascular tissue. It

then begins to suck out the phloem from the vein of vascular tissue. This behavior also

exposes them to the systemic insecticides inside the plant that were absorbed through

seed treatment or topical applications.

Aphids quickly became the limiting factor in canola production in the years following

its introduction, and canola could not be grown without aphid management (Crop profile

for Oklahoma canola 2007). Nicotinoid seed treatments (clothianidin and imidicloprid)

became available and early spring sampling protocols and economic thresholds for aphids

were developed (Giles et al. 2009, Royer and Giles 2010; Fig. 2). Seed treatments

suppressed aphid populations long enough to allow growers to delay foliar insecticide

sprays until late February or early March. Late season economic thresholds have yet to be

established, but well-timed insecticide applications during severe aphid pressure can

preserve up to 70% of potential yield. However, it is likely that late-spring pesticide

applications reduce populations of beneficial insects that inhabit flowering canola, which

could increase the need for more applications. Honeybees in particular face the potential

of detrimental effects of persistent, broad-spectrum insecticides. Thus, the introduction of

new, more species-specific insecticides (flonicamid and sulfoxaflor) will be an important

factor in the success of the agroecosystem.
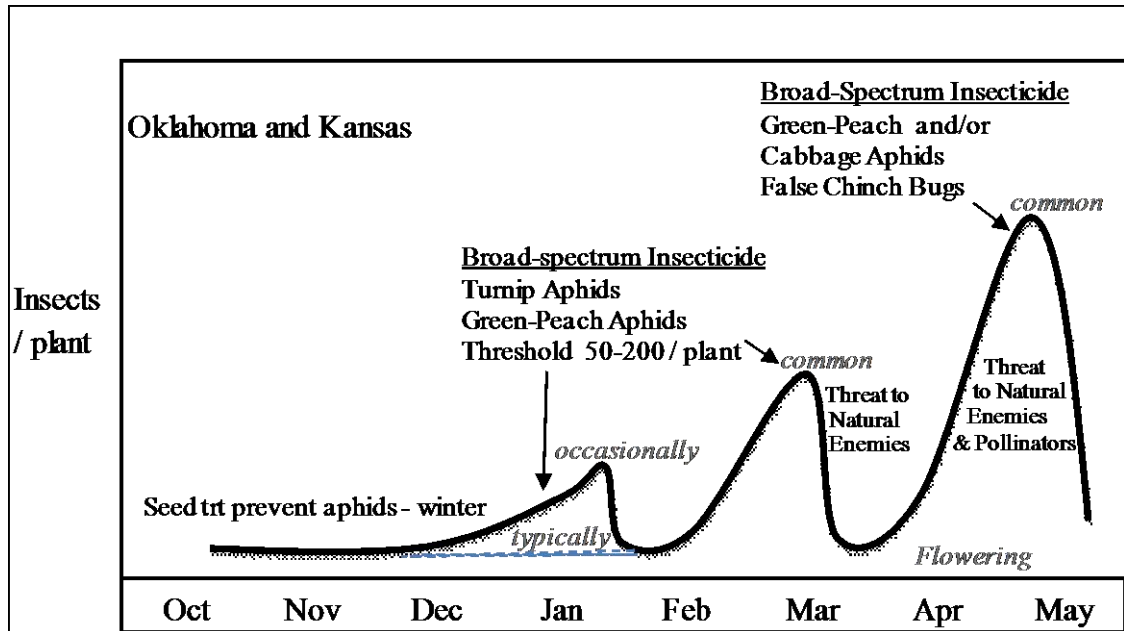
Figure 2. Current pest dynamics with broad-spectrum insecticides in winter canola
(Royer and Giles, 2010).

The application of broad-spectrum insecticides in the above management strategy

results in a swift resurgence of aphid populations in April and May. Table 1 below shows

an average outbreak in the spring, demonstrating the short term effectiveness of

pyrethroid treatments and how the reduction of natural enemy populations can allow

aphids to resurge (K. L. Giles, unpublished data).

Table 1. Number of turnip aphids per plant in Perkins Oklahoma, 2006. Comparison of untreated canola versus canola treated with synthetic pyrethroids.

Treatment Formulation	Rate Lb (AI)/acre	Pre-treatment (23 Feb)	7 DAT (2 Mar)	22 DAT (17 Mar)
Untreated	0	97.4	166.3	257.0
Mustang MAX0.8EC	0.025	82.3	20.8	83.3
Capture 2EC	0.041	124.2	1.2	56.3
Proaxis 0.5EC	0.015	105.3	2.4	110.0
Warrior	0.03	91.4	0.6	53.5

Ninety percent of fields are annually treated with broad-spectrum pyrethroid insecticides (Franke et al. 2009) to control high densities of aphids, which coincides with the time when natural enemy and pollinator populations are high (particularly during flowering). As the only flowering plant available in the early spring, canola fields attract beekeepers' and feral bees from nearby nesting sites in pastures and field margins (Stanley and Stout, 2014), while aphid infestations bring in insect predators. Current pest management strategies using pyrethroid insecticides are therefore not sustainable due to the direct toxicity to beneficial insect species.

Broad-spectrum insecticide sprays have greatly increased the synthetic inputs into the areas previously dominated by winter wheat and pasture, as less that 16% of wheat fields are annually treated (Giles et al. 2003, NASS 2005-2009, Giles and Walker 2009). The past century has seen mainly low-input systems in the southern US (Epplin et al.

1998, Crop Profile for wheat in Kansas 1999, Smith and Anisco 2007, Crop Profile for

wheat in Oklahoma 2005, NASS 2005, NASS 2008), and the introduction and growth of

the canola industry in the area could potentially expose up to three million acres in

Oklahoma, Texas, and Kansas to pesticide treatments.

Aphid and natural enemy populations were documented in an untreated canola

field in late spring (Chown and Giles, 2006; Table 2). As with similar untreated fields in

the area, aphid densities cause nearly complete crop loss. Common natural enemies to

aphids found in the field were Coccinellidae (*Coccinella septempunctata*, *Hippodamia*

convergens, and *Coleomegilla maculata*) and aphid parasitoids (*Lysiphlebus testaceipes*

and *Diaeretiella rapae*).

Table 2. Late-spring abundance of canola insect pests and natural enemies per plant by
month and day in Perkins, Oklahoma in 2006. LB = Lady beetle. DB Moth =
Diamondback Moth.

Species	3/3	3/14	3/23	3/31	4/5	4/14	4/23	5/1	5/8	5/23	5/30	6/7
CA	15	58	28	74	430	1083	3520	4325	480	20	4	
GPA	3	19	5	32	12	40	343	475	650	75	8	
TA					2	3	54	5	30	45		
DB Moth								1	1			
C. sept.			1		6	276	875	680	115	8	4	2
H. con.						1	15	32				
C. mac.						1	15	65	2			
LB Eggs					2	2						
LB Larvae					4	140	684	800	6			
LB Pupae						4	280	275				
Parasitoid Mummies		1			3	475	1260	1750	120	4		

In a preliminary study in 2011-2012 (Giles, unpublished data), 16 winter canola ecosystems were monitored for common natural enemies, lacewings and lady beetles (*Chrysoperla carnea, H. convergens,* and *C. septempunctata*). The study showed that prior to insecticide applications, there are significant numbers of natural enemies in the agroecosystem. Studies have recently begun to quantify the roles of natural enemy populations in pyrethroid-treated canola (Mccornack et al.), but no data to date describes the potential pest suppression of natural enemies in winter canola.

Lady Beetles and Biocontrol

There are 453 aphidophagous lady beetle species in North America (Gordon 1985), common in most habitats and capable of long distance flights (Hagen 1962). Species are variable in size, shape, and color but typically have a red-orange color and black spots on their elytra. The native species *H. convergens* and the exotic species *C. spetempunctata* are considered the most common lady beetles found in the Southern Great Plains (Teetes et al. 1973, Elliott et al. 2006). Aphids in crop systems are a primary source of prey for lady beetles, making them beneficial in fields with frequent aphid outbreaks (Kring et al. 1985, Rice and Wilde 1988, Elliott et al. 1996, Jones 2001).

Lady beetles lay clusters of 5-50 eggs distributed on the undersides of leaves (Honěk 1996). Larvae are known to cannibalize their siblings before migrating away from the hatching site to search for food. Cannibalism and interspecific predation are common phenomena among lady beetle species and potentially enhance survival rates when prey is scarce (Agarwala and Dixon 1992, Hodek 1996, Obrycki et al. 1998, Snyder et al. 2000). Larvae molt three times in a 14-day period and spend several days immobile as a fourth instar, or "prepupa", before pupating (Hodek 1996). Larval and pupal stage

durations vary due to differences in ambient temperature and prey and water availability (Honěk 1996, Michels and Behle 1991, Phoofolo et al. 2007, Royer et al. 2008).

After pupation, *H. convergens* adults have a pre-ovipositional period lasting from 6 to 12 days depending on the quality and availability of food (Gutierrez et al. 1981). *Hippodamia. convergens* lays an average of 20 eggs per day and may lay hundreds of eggs in a lifetime, able to produce multiple generations within a single year (Honěk 1996). Adults can overwinter, or diapauses, in protective vegetation, the duration of which is largely determined by prey availability, and are able to feed on other food sources such as pollen when aphid prey is limited (Hagen et al. 1976, Hemptinne and Desprets 1986). Pollen and nectar consumption are also known to be vital to successful diapause and reproduction (Hagen 1962).

The quantity and quality of prey largely determine lady beetle survival to adulthood (Hodek and Honěk 1996, Agarwala 2008). Slower developmental rates, increased pupal mortality, and decreased ovipositional capacity are a result of food stress (Srivastava 2003, Royer et al. 2008, Takizawa et al. 2000). The delay in development may be an adaptive strategy designed to attempt to meet the nutritional requirements or body weight needed for successful molting (Davidowitz et al. 2003, Phoofolo et al. 2008). However, final instar stages exposed to very limited diets are generally unable to recompense starvation effects (Baumgaertner et al. 1981).

It has been determined that the nutritional suitability of a lady beetle's prey is influenced by the prey's host plants which in turn affects the success of lady beetle populations (Starks et al., 1972; Rice and Wilde, 1989; Kareiva and Sahakian, 1990; van Emden and Wratten,1990; Obrycki and Kring, 1998; Bottrell et al., 1998). The toxicity of

prey resulting from the biochemical nature of their host plant may have adverse effects on predators, including increased mortality and decreased fecundity and growth rates (Kareiva and Sahakian, 1990; Power, 1992; Hodek, 1993; Hodek and Honek, 1996; Bottrell et al., 1998). Monitoring aphid nutritional value as food is important when observing tritrophic systems because aphids store different nutrients from plants based on the host plant species and cultivar (Dillwith et al., 1993). The physiological status of host plants determine aphid suitability as a food source (Klingauf, 1988; Srivastava, 1988; and Dixon et al., 1998), as well as the nutrition and ingested biomass requirements of the lady beetles that consume them (Ferran 1978; Eubanks and Denno 2000). Lady beetles have been shown to avoid prey treated with certain pesticides: for instance, lady beetles had significantly reduced consumption of aphids sprayed with lambda-cyhaothrin, a pyrethroid, resulting in increased survival of those individuals (Thornham et al., 2007).

Lady beetles, as generalist predators, are able to select their prey items to maximize a favorable nutrient intake. This dietary self-selection, as coined by Waldbauer and Friedman in 1991, may result in lady beetles choosing not to feed on toxic aphids in field conditions if other prey is available, reducing their effectiveness at biological control. Lady beetles use environmental clues to find aphid prey, making them efficient foragers.

Olfactory cues have been shown to play a major part in the searching behavior of lady beetles (Obata 1986, 1997, Ponsonby and Copland 1995, Zhu et al. 1999, Ninkovic et al. 2001). Following an initial encounter with a prey item, lady beetle adults and larvae often remain in the area to continue searching, turning side to side, allowing them to successfully find more prey. This is an adaptive behavioral response to the tendency of

aphid populations being clumped in the landscape, instead of being evenly distributed. Newly hatched lady beetle larvae must find food within about 30 hours or they will die of starvation, making oviposition sites an important part of lady beetle survivability. Thus, lady beetles produce more eggs as their rate of aphid consumption increases, allowing them to lay eggs in areas of high aphid density (Evans, 2003).

In addition, lady beetle larvae are highly mobile and are known to search actively and continuously for prey as long as they have the energy required to do so (Banks, 1957). Several studies have shown that predators respond to odors released by herbivore-damaged plants to navigate towards potential prey (Vet and Dicke 1992, Drukker et al. 1995, Bruin et al. 1992, 1995, Dicke, 1999, Sabelis et al. 1999, Francis et al. 2004). Lady beetles have been shown to navigate towards leaves being fed upon by aphids and towards leaves that had been previously fed upon (Jamal and Brown, 2001). Lady beetle adults are highly mobile, but become less active when encountering areas of high aphid populations.

The success or failure of a lady beetle attacking and feeding on an aphid prey depends on many individual factors, such as the predator's hunger level, age, and genetic characteristics, as well as the presence or absence of inter- and intra-specific competition, the shape and structure of the plant host, the species of aphid prey and its defenses, and the interference by protective ants, which can defend aphid colonies against lady beetles of all size and species (Ferran and Dixon, 1993).

Many different species of lady beetles feed on aphids, and combined with other predators and parasitoids, can effectively maintain aphid populations below economic thresholds in many crop systems. Lady beetles have been utilized in classical biological

control in the United States since 1889 to control a wide range of pest species. Aphidophagous lady beetles have been widely successful at suppressing aphid populations in field, laboratory, and greenhouse conditions (Hodek and Honek, 1996) and continue to be a significant mortality factor of aphids. In an experiment by Snyder and Ives in 2003, the removal of lady beetles from a multiple control factor system on pea aphids resulted in an immediate increase in aphid population growth. Adult seven-spotted lady beetles (*Coccinella septempunctata L.*) can eat an estimated maximum of 277 soybean aphids per day, depending on prey densities, temperature, and other environmental factors (Xue et al., 2009). Lady beetle adults are highly mobile and thus able to aggregate rapidly in response to heavy aphid infestations. Aphids are considered an essential food item for lady beetles, which are generalist predators and many species also feed on pollen and fungi (Hodek and Honek, 1996). Lady beetles may be best utilized in an agricultural setting in combination with human intervention because lady beetles cannot provide long-term pest population regulation, especially in the case of an aphid outbreak (Sun et al., 2000).

Peterson et al. in 2009 proposed the consideration of every mortality factor affecting an organism to create an accurate picture of how just one mortality factor can affect its population dynamics. They re-analyzed dozens of insect life tables and came to the conclusion that many mortality rates by predators, pathogens, and parasitoids could be replaced by abiotic factors, resulting in the overestimation of mortality rates attributed to biological control. They suggested that the stability and degree of protection in the environment greatly influences the magnitude of irreplaceable mortality, which is defined

as the portion of mortality that cannot be replaced by another cause. Essentially, the stability of the environment will affect the success of natural enemies.

Canola and Aphid Defense

Canola (*Brassica napus*), as well as other members of Brassicaceae, can synthesize glucosinolates that are utilized in a defense system against herbivore feeding (Wang et al., 2011). Canola plants produce glucosinolates and myrosinase but keep them spatially separated in the plant cells until the cells are damaged. Then the compounds are combined as needed to produce toxic products, effectively termed a "mustard oil bomb", to discourage further feeding by herbivorous insects (Ratzka et al. 2002).

Cabbage aphids and other insects have evolved the ability to feed on plants containing these compounds, and sequester glucosinolates into their own bodies. They then synthesize their own myrosinase compounds (Jones et al. 2001, 2002; Pontoppidan et al. 2001; Bridges et al. 2002; Husebye et al. 2005) and can release isothiocyantae, a product of glucosinolate hydrolysis, in defense against natural enemies (Francis et al. 2004, Franics et al. 2001). Unborn aphid young have been documented to already contain myrosinase compounds, and when attacked by lady beetles, nymphal and adult aphids have been documented to release isothiocyantaes and an alarm pheromone (Kazana et al. 2007).

Lady beetle larvae exposed to aphids fed on a diet high in glucosinolates failed to reach adulthood (Francis et al. 2001), and in another experiment, had a greatly increased mortality compared to a control (Kazana et al. 2007). This could be a reason that lady beetle populations remain relatively low in canola compared to those in wheat in the Great Plains, with wheat containing more than twice the number of lady beetles in canola

(Giles, unpublished data). Jessie in 2009 (unpublished data) found that a diet of certain aphid species fed on winter canola resulted in lower adult body weight of *H. convergens* lady beetles, but did not significantly affect developmental times, and that green peach aphids on a canola diet should be considered a high-quality food source of the beetles.

Canola fields may be a sink for natural enemies: predators from wheat and other crops may be able to move in to canola fields, but once there are faced with unsuitable prey populations. The combined result of annual pyrethroid applications unsuitable prey likely result in low levels of biological control of pests, but there is little research preformed on this new area of interest since canola is a relatively new crop to the Great Plains.

Pesticides and IPM

One goal of an Integrated Pest Management program is to suppress pest species in sustainable, environmentally friendly ways by utilizing chemical, cultural, and biological forms of control. However, achieving compatibility between chemical and biological control is difficult: many pesticides on the market today are broad-spectrum, meaning they have equally devastating effects on both the pest and natural enemy populations. To reach the goals of IPM, pesticides must be selected to attain the highest improvement of natural enemy action (Sterling, 1984). In order to improve and sustain biological control, pesticides used must be low in toxicity to natural enemies and support their longevity. Insecticides have both direct (i.e. mortality from contact with the pesticide) and indirect (i.e. consuming treated prey) toxicity to insects (Debach and Rosen, 1991). Surviving natural enemies can be affected by a reduced food source, declines in their longevity and fecundity, and a tendency for adults to disperse from a treated area due to the

insecticide's ability to repel the pest populations (Newson, 1974). Annual and perennial crops are considered disturbed environments due to constant human activity and inputs. This type of environment makes successful biological control difficult because of the disruptions to the ecosystem (DeBach 1964). Traditionally, it is believed that specialist biocontrol agents are more successful at suppressing pest populations because their life histories more closely resemble that of their prey (DeBach and Rosen, 1991). However, generalist predators such as lady beetles still may provide adequate biological control in agricultural settings because of their ability to survive on other hosts (Doutt and DeBach, 1964; Miller, 1977). Thus, because of their abundance in the Southern Plains (Hodek and Michaud, 2008), lady beetles such as *C. septempunctata* show a promising opportunity in canola for increased aphid suppression if natural populations are maintained by using selective pesticides.

Flonicamid

Flonicamid (Figure 3) is a novel chemical introduced by Ishihara Sangyo Kaisha, Ltd to the world market in 2005 for the purpose of killing aphid and thysanopteran pests. Flonicamid belongs to Group 9C in IRAC's (Insecticide Resistance Action Committee) Mode of Action table (IRAC MoA Classification Scheme). Group 9 compounds are pyridinecarboxamides, a relatively new and largely unstudied class of chemicals not previously utilized as insecticides. Also in group 9 is pymetrozine (in products such as Fulfill and Endeavor), which is a similarly a feeding inhibitor. Flonicamid is available to use on several crops including alfalfa, cotton, and avariety of fruits and vegetables. Carbine, Beleaf, and Aria are commonly used products containing %50 flonicamid as the active compound.
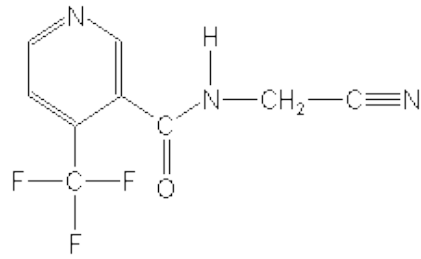
Figure 3: Chemical structure of flonicamid

Flonicamid acts as a rapid feeding inhibitor against insects with piercing-sucking mouthparts, including aphids, thrips, scale insects, mealybugs, leafhoppers, and whiteflies, by preventing the stylet from penetrating the leaf surface (Morita et al. 2007, Cho et al. 2011, Jansen et al. 2011). Aphids affected by flonicamid still attach the head of the proboscis to the leaf surface, but are unable to salivate and feed (Morita et al., 2007). The mode of action of flonicamid is through the nervous system, but its exact pathway is not yet solidly identified. There is evidence that flonicamid acts through blocking the potassium A-type channel (Staetz et. al, 2006). The blocking of potassium channels results in a loss of nervous system control, which leads to stylet mouthparts becoming flaccid and unable to probe into plant tissue. The graphs below (Figure 4) demonstrate the result of flonicamid treatment on *Myzus persicae* nymphs and how flonicamid first inhibits feeding and then results in mortality.
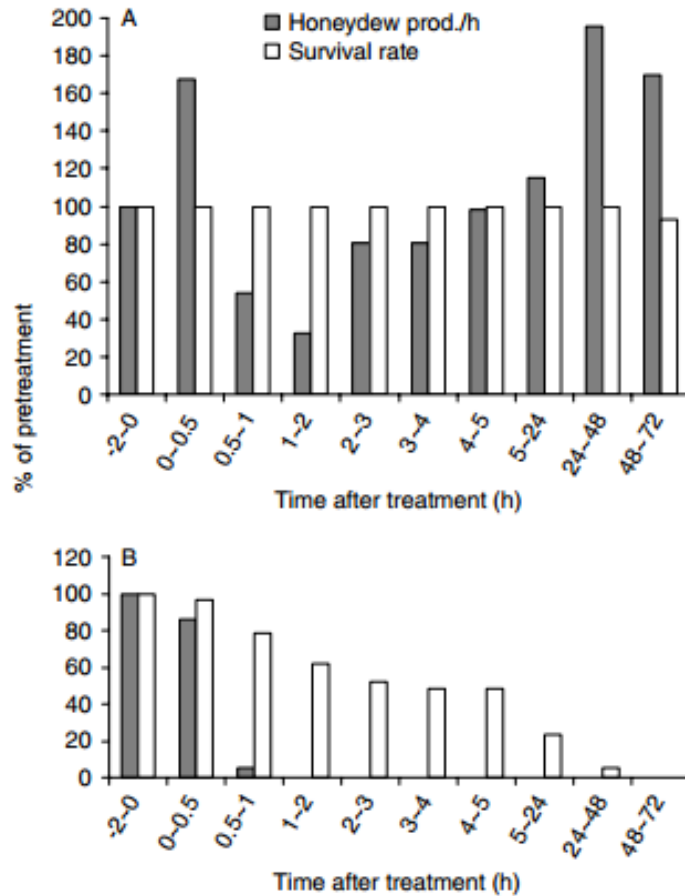
Figure 4. Effects of flonicamid on honeydew production and survival rate of first instar nymphs of *Myzus persicae*: A. control aphids; B. aphids treated with flonicamid at 100 mg AI $^{L-1}$.

After short term exposure, aphids exhibit severely reduced feeding and mortality due to starvation, and are unable to recover feeding ability before they die. In the experiment by Morita et al. in 2007 (Table 3), they found that aphids affected by flonicamid took up to 72 hours to drop from the leaves of host plants, but honeydew production showed a rapid decline after just a half hour of the aphids being sprayed. This indicates that flonicamid quickly inhibits feeding and then aphids die of starvation, with the LC_{50} for key aphid species being 0.64 and 2.01mg AI L^{-1}. Also, the nymphs produced by treated aphids also have high mortality rates, even though they had no direct contact

with the insecticide. Flonicamid is successful against both adult and nymphal aphids, as

Mortia et al. shows in the same experiment on wingless adults.

Exposure period (h)	Mortality of adults (%)	Number of nymphs produced per ten adults	Mortality of nymphs produced (%)
1	35	64	74
3	60	45	90
Control	10	100	0

ª Assessments were made 72 h after the end of each exposure period.

Table 3. Effects of short-term exposure to flonicamid at 100mg AI L-1 on wingless adults of *Myzus persicae*.

Flonicamid shows a strong potential for use in Integrated Pest Management systems due

to its low environmental impacts: flonicamid has been shown to have little toxicity to

natural enemies and pollinators (Jansen et al. 2011, Maienfisch et al. 2012). Many studies

have been conducted to determine the effects of numerous insecticides against lady

beetles, and toxicity varies widely by species and classes of insecticides (Kaakeh, 1996).

Flonicamid, especially when compared to pyrethroid and neonicitinoid pesticides, has

been found to have very low toxicity to many species of lady beetles (Jalali and Leeuwen,

2009; Cloyd and Dickinson, 2006; Jansen et al., 2011). Flonicamid has not been shown to

exhibit cross-resistance to other classes of insecticides and has low toxicity to beneficial

insects, including pollinators and parasitoids (Nieto and Simonetta, 2008; Cloyd and

Dickinson, 2006). Flonicamid insecticides offer a safer, more environmentally-friendly

approach to pest control in many different crops. Insects are not yet resistant to the

unique mode of action, and they pose no known threat to pollinators or natural enemies.

The toxicity of pesticides to lady beetles can be measured in direct mortality or indirect

(sub-lethal) mortality, which encompasses delays in population growth, individual

growth rate, fertility, and other factors that decrease the population. All of these aspects must be accounted for to determine if a pesticide is safe for natural enemies (Stark et. al, 2007).

In certain products, flonicamid exhibits translaminar activity on crops plants, meaning it can penetrate the leaf tissue and form a reservoir of active ingredient that can provide sustained residual activity. Therefore, getting full spray coverage is not necessary because the pesticide can move systemically through the plant, effectively reaching pests that feed on the undersides of leaves, like aphids. The exact mechanism of aphid uptake is unclear in the available literature: aphids could consume flonicamid from treated plants by ingesting it with their proboscis, or be affected by coming in contact with sprayed leaf surfaces through other body parts, such as the tarsi. Basically, it is unknown if aphids must directly consume flonicamid, or if it can be absorbed into the cuticle.

As previously stated, direct contact with flonicamid is reportedly safe for beneficial insects, but the effects of eating treated prey are unknown. In addition, the long-term effects of flonicamid and the effects of application methods (seed treatment versus foliar applications) on a lady beetle-aphid system have not yet been tested.

Sulfoxaflor

Sulfoxaflor has a similar function as the neonicitinoids (Culter et al. 2012) but belongs to a new class of insecticides called sulfoxamines (Zhu et. al 2011). Sulfoxaflor is successful at suppressing sap-feeding pests including aphids (Kerns et al. 2010, Annetts et al. 2012) and was approved for use in June 2013. It does cause low levels of mortality to honeybees when they are exposed to dry residues on plant material or directly exposed, but has no long term effect on the population (Jeschke et al. 2012).

Figure 5 (Giles, unpublished data) demonstrates the potential numerical response in a system containing natural enemies and aphids treated with narrow-spectrum insecticides like flonicamid. The slow decline of aphid prey supports natural enemy populations that are able to survive until aphids recolonize the system in late spring, and can also feed on the abundant pollen supply. Bushong et al. in 2012 examined the net benefits to producers of using narrow spectrum insecticides. They found that positive net gain was affected by the ability to control pest populations, and narrow-spectrum insecticides may allow for fewer insecticide sprays due to natural enemies preventing population resurgence.
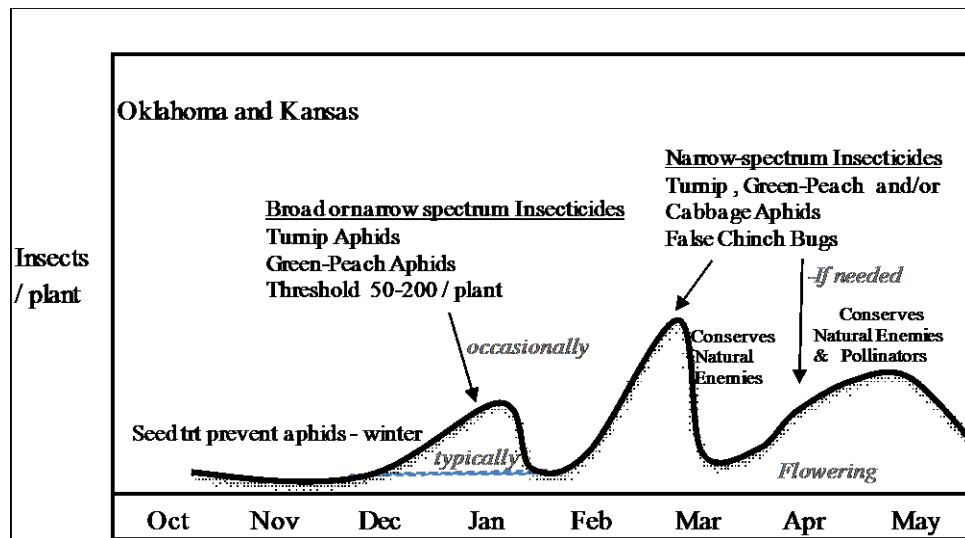

Figure 5. Potential dynamics with narrow-spectrum insecticides in winter canola.

Conclusions and Research Needs

Lady beetles have been shown to successfully control aphid populations (Abd-Rabou, 2008) in many different agricultural media and can produce a secondary method of mortality of aphids if their natural populations are augmented or left unharmed in the

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field. The use of a low-toxicity insecticide in combination with natural beetle predation could result in successful, long-term aphid suppression and reduce the risk of resistance developing due to aphids facing two means of mortality. Incorporating multiple mortality factors to control pests is a key component of integrated pest management practices which promote sustainable agriculture. Canola crops have the potential to produce toxic aphid prey and therefore may be unsuitable habitat for lady beetle populations to provide biological control. The addition of a feeding suppressant such as flonicamid may influence lady beetle success by inhibiting the chemical defense system of aphids utilizing the glucosinalates in canola. If aphids are unable to feed, they may not be able to produce the toxic "mustard bomb" effect, which may increase their palatability for lady beetles. In addition, due to the delayed die-off of aphids treated with flonicamid, lady beetle survivorship may be extended in that the food source persists for a longer period of time.

Canola producers in Oklahoma and the southern Great Plains are limited to a small list of effective pesticides for aphid control (Royer and Giles 2012, 2013 OSU Extension Agents' Handbook).

To ascertain if these new narrow-spectrum insecticides are compatible with current IPM programs, their direct and indirect effects on beneficial insects must be determined. We must quantify the effects of these chemicals on the development, survival, and reproduction of natural enemies. If these new insecticides have little to no effects on the population dynamics of beneficial insects, they would be ideal as an integrated pest control option for producers.

CHAPTER III

MATERIALS AND METHODS

Aphid Colonies. Winter canola (cv. 'Wichita') plants were planted weekly in

potting soil in 30-cm-diameter pots and maintained in a growth chamber at 24°C and a

photoperiod of 16:8 (L:D). Faba bean (*Vicia faba* cv. 'Windsor') were planted in 15-cm-

diameter pots with a 50:50 mixture of potting soil and fritted clay with Osmocote® 15-9-

12 pellet fertilizer and watered as needed. Winter canola and faba plants were transferred

to aphid colonies once they reached 25 and 7 days in age, respectively.

A colony of green-peach aphids was established from individuals collected from

winter canola fields in Central Oklahoma. Green peach aphids are a highly suitable food

source for both *H. convergens* and *C. septempunctata* (Jessie, 2013). The colony was

maintained in a growth chamber at 24°C and a photoperiod of 16:8 (L:D). Pots and plants

were replaced weekly as needed. Stock colonies of pea aphids reared on faba bean were

maintained in large, single-walled mesh boxes kept at 24°C and a photoperiod of 16:8

(L:D). Pea aphids were collected daily to prevent plant death and fresh plants were added

to the colonies weekly. All winter canola aphids used in experiments were transferred to

the individual experimental unit arenas (5-cm-diameter cups).

Ladybeetle Colonies. Adult *H. convergens* and adult *C. septempunctata* were collected from winter canola fields in central Oklahoma and transferred to 0.25-liter cardboard containers topped with fine, nylon mesh lids. All adult lady beetles were maintained in environmental growth chambers at 24°C and a photoperiod of 16:8 (L:D) and were provided daily with an unlimited supply of fresh pea aphids, a moistened cotton ball, and a honey-wheat-yeast mixture (Planet Natural Garden Supply©, Bozeman, MT). Egg clutches were collected daily and stored as needed in a growth chamber at 10° C to suspend hatching before use in experiments.

Individual Feeding Trials. The first experiment was designed to evaluate the suitability of aphids treated with narrow-spectrum insecticides on the survival and development of two common Coccinellids found in winter canola. Forty-eight hours prior to initiating feeding trials, three separate pots of heavily infested canola plants (containing green-peach aphids) were sprayed with one of three treatments. A medical grade atomizer was used in application of all treatments. The first two pots were sprayed with flonicamid and sulfoxaflor (mixed with water) at field rates based on pot size and label instructions (Beleaf and Transform insecticide labels) for aphids on canola (Beleaf at 2.8 oz/acre and Transform at 0.75 oz/acre), and the third pot was sprayed with an equal volume of water (control). Sprayed pots were isolated in separate plastic tubs for twenty-four hours in growth chambers.

Upon hatching, first instar ladybeetle larvae were individually placed in separate plastic 5-cm-diameter cups, and provided daily with an unlimited diet of freshly collected green-peach aphids from one of the three treated pots. Leaves bearing green peach aphids were placed directly in feeding cups. Green peach aphids from treated pots were

used for forty-eight hours without being retreated. Surplus treated aphids on canola leaves were stored in Ziplock bags in a refrigerator (4° C) and used during larval development. Using this timescale, a new set of pots was sprayed every forty-eight hours. All diet/insecticide treatments were replicated with twenty separate individuals of each species. Mortality, molting, pupation, and emergence were recorded systematically every 24 h for each larva. Upon adult emergence body length and width were determined and used to calculate elliptical body area [$\pi \times$ ½ (body length) $\times$ ½ (body width); Obrycki et al. 1998]. Adults were preserved in alcohol and labeled specimens were deposited as voucher specimens in the OSU K.C. Emerson Insect Museum.

Microcosm Competition Trials. The second experiment was designed to evaluate survival and development of competing Coccinellidae larvae on insecticide treated plants with aphids. Separate pots of heavily infested canola plants were sprayed with the three treatments as described above. Individual plants were removed from the pots and transplanted into 10-cm-diameter pots filled with potting soil and topped with a layer of sand. A plastic cylinder top covered with a mesh cap was placed over the plant and pressed into the soil to seal off each pot. After 24 h four newly hatched 1st instar ladybeetle larvae were introduced into each pot. For each insecticide treatment the following 4-larvae intraspecific and interspecific competition scenarios were introduced into individual microcosms: 4 *H. convergens* (5 replications), 4 *C. septempunctata* (5 replications), and 2:2 *H. convergens*:*C. septempunctata* (2 replications). Each day, aphid presence/absence was noted and larvae were counted and stage identified based on presence of exuviae and size of larvae. Data were summarized for comparisons every three days.

Statistical Analysis. Statistical analyses were performed using SAS version 6.12 (SAS Institute. 1996). The significance level chosen for all analyses was $p = 0.05$. Development times and adult elliptical body area (mm^2) of *H. convergens* and *C. septempunctata* were compared among treatments by ANOVA (PROC MIXED; LSMEANS comparisons). Cumulative of larval, pupal, and larval+pupal survival ratios and female:male ratios were compared among and between treatments using a chi-squared analysis (PROC FREQ; Fishers Exact Test).

CHAPTER IV

RESULTS AND DISCUSSION

Individual Feeding Trials

Survival and Female:Male Ratios. Overall and for both predators, survivorship ratios were similar between the control and flonicamid diet treatments further suggesting that this compound is an ideal tool for integrated control in winter canola. Cumulative survival ratios of *C. septempunctata* were statistically different at each larval and pupal stage ($\chi^2 > 10$, $p < 0.007$), and were primarily influenced by reduced survivorship associated with sulfoxaflor treated aphids (Figure 6).

Figure 6: Effects of diet treatments on *C. septempunctata* cumulative survival ratio at each larval stage and proportion female.
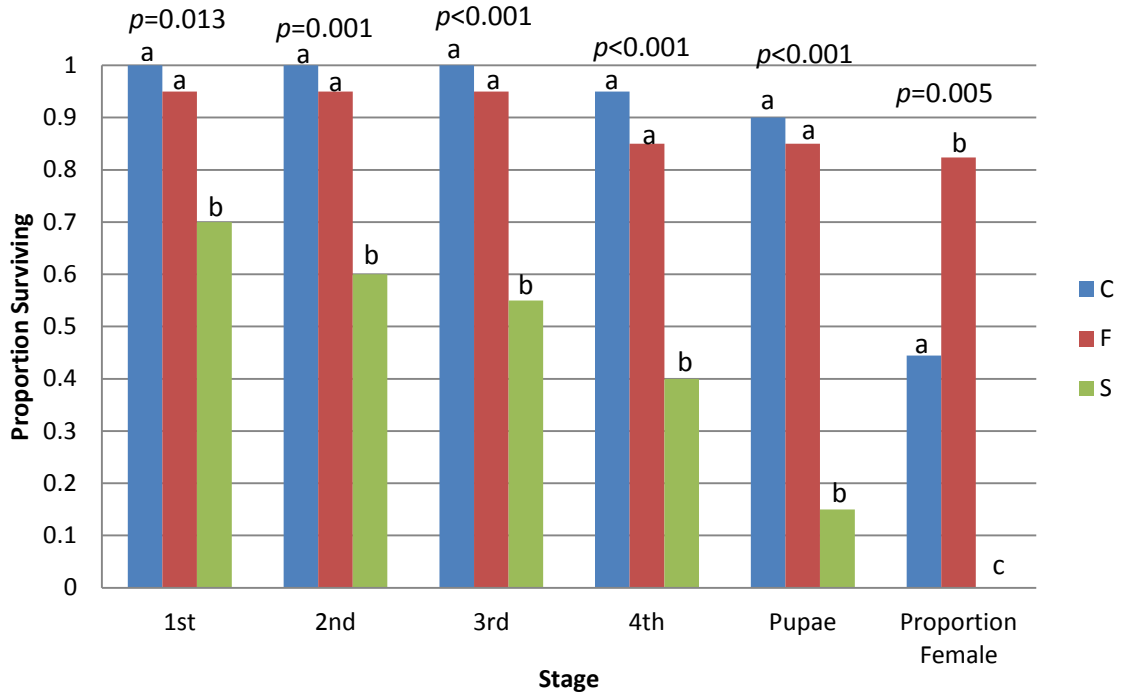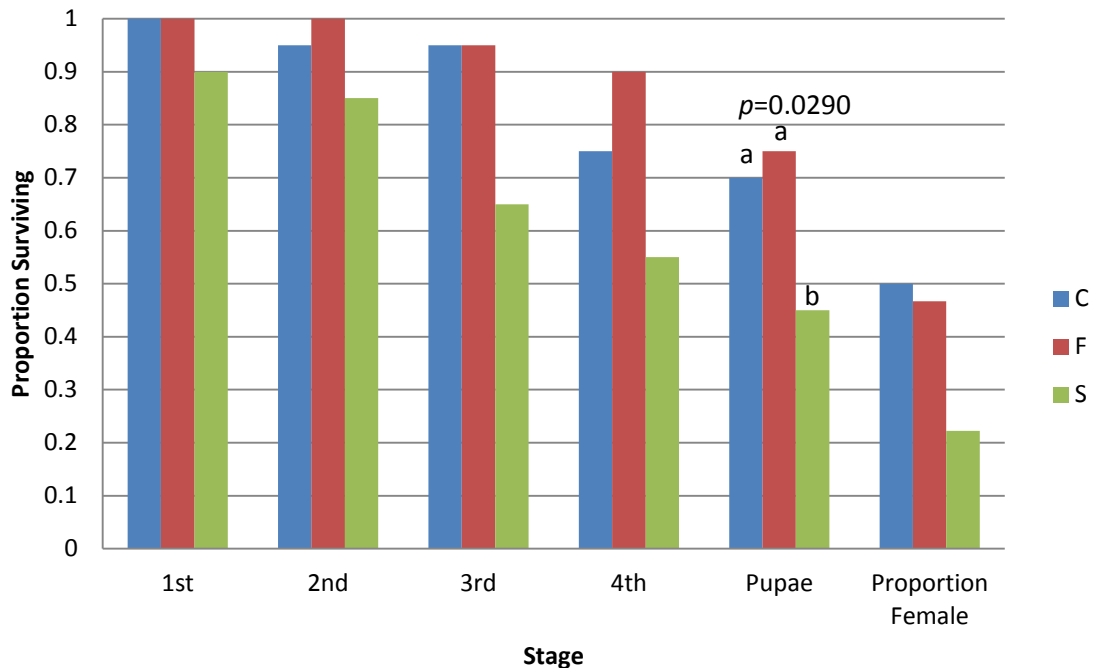

[a] Experimental treatments were water control (C), flonicamid (F), and sulfoxaflor (S) at field rates.
Bars with the same letter are not statistically significant.

Thirty percent of larvae died during the first instar, and only twenty percent survived to the pupal stage when supplied with sulfoxaflor treated aphids. Clearly, sulfoxaflor treated aphids reduce survivorship for both predators, and the impact is quite severe for *C. septempunctata*.

Despite a clear trend in reduced cumulative survival ratios (alive:dead) for *H. convergens* fed sulfoxaflor treated aphids, no significant differences among treatments within larval stages was observed (Table 4, Figure 7).

Figure 7: Effects of diet treatments on *H. convergens* cumulative survival ratio at each larval stage and proportion female.
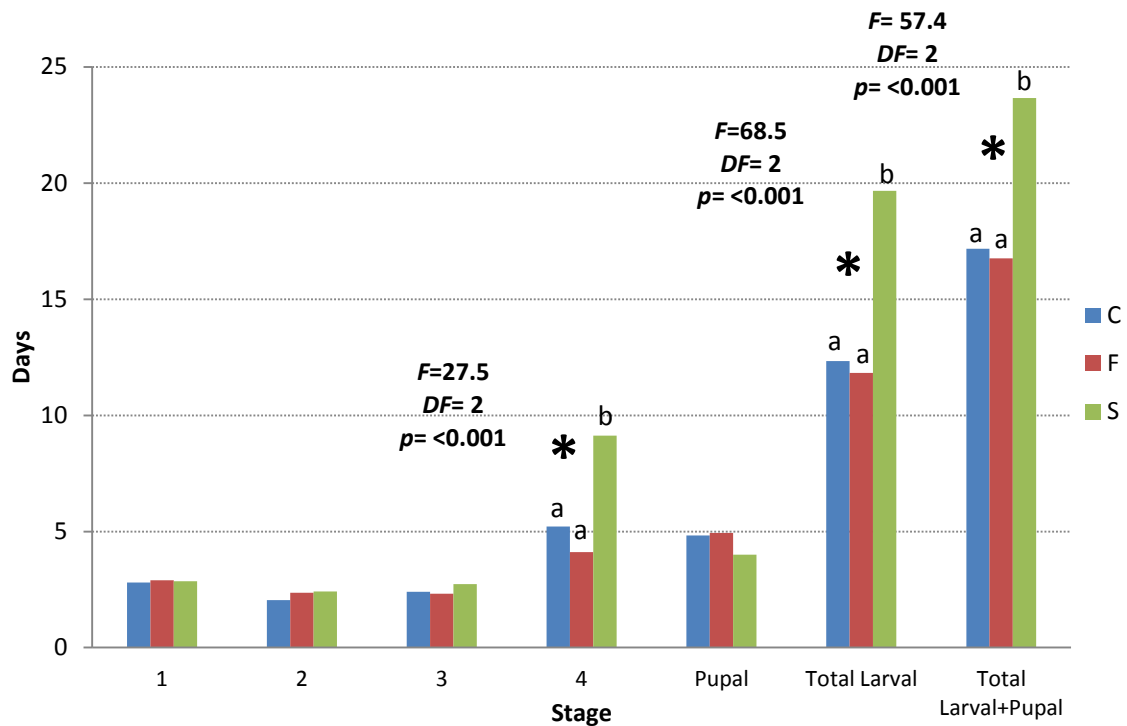

[a] Experimental treatments were water control, flonicamid, and sulfoxaflor at field rates. Bars with the same letter are not statistically significant.

However, incorporation of pupal mortality (total preimaginal survivorship) resulted in significantly reduced survivorship for *H. convergens* fed sulfoxaflor treated aphids (Table 2). Based on survivorship findings, sulfoxaflor is clearly more toxic to Coccinellids than flonicamid, but could still be an important integrated control tool in canola fields. Compared with synthetic pyrethroids, such as lambda-cyhalothrin, which are highly toxic and rapidly kill immature Coccinellidae (Jalali and Leeuwen 2009, Jansen et al. 2011), a decreasing proportion of beetle larvae could continue to feed on sulfoxaflor treated aphids that are dying and this combined mortality may reduce the chance of aphid resurgence in winter canola fields.

Without assays to document the presence of male-killing bacteria (Majerus and Hurst 1997), it is not fully possible to interpret the effects of diet treatments on sex ratios for Coccinellidae (Table 5, Figures 6 and 7). It is quite interesting to note, however, that flonicamid treated aphids resulted in a much higher percentage of *C. septempunctata* females (80% female) than the control. Additionally, the fact that there were no surviving *C. septempunctata* females in the sulfoxaflor treatment indicates a potential severe effect of this insecticide on fitness and population dynamics in canola fields. The proportion of female *H. convergens* was lower on sulfoxaflor treated aphids, but not statistically different from control and flonicamid treatments (χ^2=5.7, *df*=2, *p*=0.060). Further studies are clearly needed to isolate the effects of flonicamid and sulfoxaflor on Coccinellid sex ratios and reproductive potential in treated canola fields.

Development. For both *H. convergens* and *C. septempunctata*, development times were significantly different for a few larval stages among the three diet treatments (Figures 8 and 9; Table 4). For both species, total larval and total larval + pupal development times were not different between the water control and flonicamid (*C. septempunctata p*=0.173, *p*=0.263, *H. convergens p*=0.222, *p*=0.342).
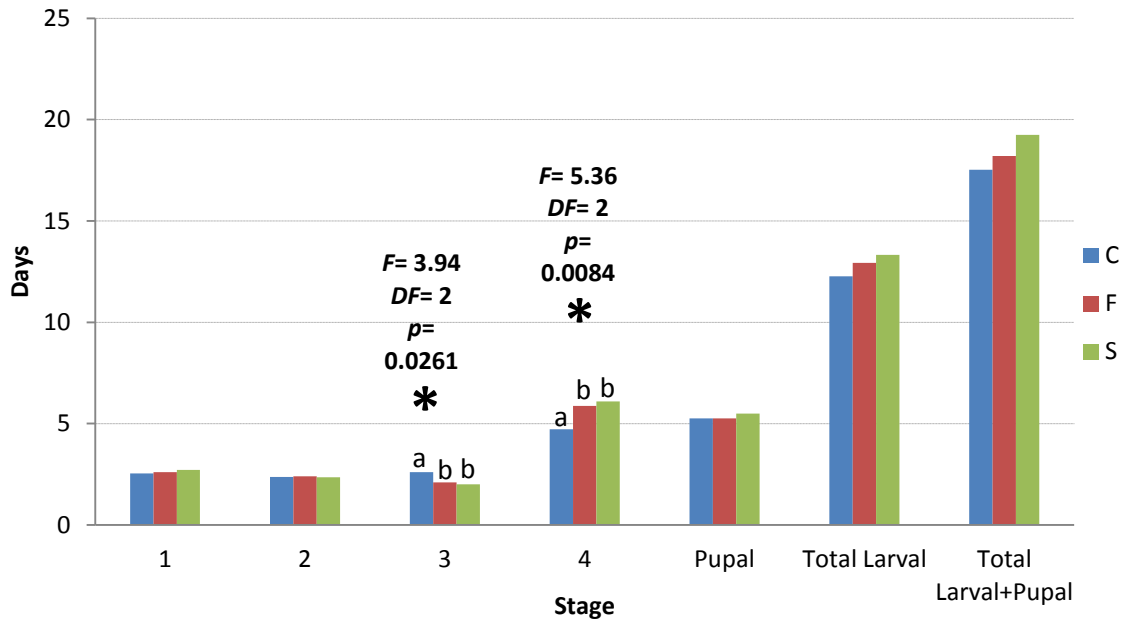
Figure 8: Mean duration of immature stages of *C. septempunctata* feeding on treated[a] *M. persicae* at 24°C.

[a] Experimental treatments were water control, flonicamid, and sulfoxaflor at field rates. Bars with the same letter are not statistically significant.

Total larval development and larval + pupal development times for *H. convergen*s were similar among treatments. However, total larval and larval + pupal development times for *C. septempunctata* significantly increased when fed aphids treated with sulfoxaflor (*F*=68.5, *df*= 2,35 *p*= <0.001)**,** and this overall increase was primarily attributed to differences during the 4th instar (*F*=27.5, *df*=2,41 *p*=<0.001). For *H. convergen*s, 3rd and 4th instar development was observed to be significantly different among diet treatments (*F*>3.9, *df*=2,48, *p*<0.026), but these differences were not consistent; the effect of flonicamid alternated between instars (Figure 9).

Figure 9: Mean duration of immature stages of *H. convergens* feeding on treated[a] *M. persicae* at 24°C.

[a] Experimental treatments were water control, flonicamid, and sulfoxaflor at field rates. Bars with the same letter are not statistically significant.

Longer development times expose larval lady beetles to additional mortality factors (starvation, predation, pesticide sprays in the field), which ultimately could reduce their survivorship. The increased duration during the 4th larval stage for *C. septempunctata* feeding on sulfoxaflor treated aphids indicates a toxic effect which could negatively impact populations of this common predator and ultimately decrease their ability to control aphid pests. Contrarily, flonicamid treated aphids had no overall effect on total development times for either predator species (compared with a water control), thus flonicamid has the potential to be an important integrated control tool in winter canola. Based on development times alone, sulfoxaflor could also be an important integrated control tool, because *H. convergens* is the primary Coccinellid inhabiting canola fields during treatment. Indeed, during a 2011-2013 survey on 18 canola fields,

H. convergens significantly outnumbered *C. septempunctata* on sticky traps during the

spring as fields were treated for aphid infestations (C. Jessie unpublished data).

Table 4: Mean duration of immature stages (in days) of *C. septempunctata* and *H. convergens* feeding on treated *M. persicae* at 24°C.

	Control	Flonicamid	Sulfoxaflor	Ratio Comparisons		
C. septempunctata				df	F	P
1st Instar	2.80 ±0.10	2.89 ±0.11	2.86 ±0.12	2,50	0.2	0.813
2nd Instar	2.05 ±0.23	2.37 ±0.23	2.42 ±0.29	2,48	0.7	0.518
3rd Instar	2.40 ±0.17	2.32 ±0.17	2.72 ±0.23	2,47	1.1	0.351
4th Instar	5.21 ±0.37[a]	4.12 ±0.39[b]	9.12 ±0.56[c]	2,41	27.5	<0.001
Pupal	4.83 ±0.22	4.94 ±0.23	4.00 ±0.55	2,35	1.3	0.295
Total Larval	12.33 ±0.26[a]	11.82 ±0.26[a]	19.67 ±0.63[b]	2, 35	68.5	<0.001
Larval+Pupal	17.16 ±0.25[a]	16.76 ±0.25[a]	23.67 ±0.60[b]	2,35	57.4	<0.001
Average Size[d] (f)	1782.82 ±110.57	2055.64 ±83.58	--	1,20	3.9	0.063
Average Size (m)	1146.61 ±78.18	1165.83 ±123.62	838.46 ±142.74	2,14	2.0	0.173
H. convergens						
1st Instar	2.55 ±0.15	2.60 ±0.15	2.27 ±0.15	2,55	0.4	0.709
2nd Instar	2.36 ±0.21	2.40 ±0.21	2.35 ±0.22	2,53	0.01	0.987
3rd Instar	2.61 ±0.16	2.10 ±0.16	2.00 ±0.18	2,48	3.9	0.026
4th Instar	4.72 ±0.30	5.88 ±0.31	6.09 ±0.38	2,42	5.4	0.008
Pupal	5.26 ±0.29	5.26 ±0.29	5.50 ±0.40	2,35	0.1	0.872
Total Larval	12.27 ±0.38	12.93 ±0.38	13.33 ±0.49	2,36	1.6	0.210
Larval+Pupal	17.53 ±0.49	18.20 ±0.49	19.25 ±0.67	2,35	2.2	0.132
Average Size (f)	1362.92 ±55.73	1181.69 ±91.01	1277.26 ±78.81	2,12	1.5	0.258
Average Size (m)	1173.67 ±83.90	1060.82 ±64.09	839.59 ±110.99	2,20	5.3	0.014

[a] Different letters following values in columns represent significant differences (p <0.05) between each treatment at each stage.
[d] Size measured in mm^2.

Adult Body Size. Because of size differences between males and females, the

effect of diet treatment was compared within sex (Table 4). Average adult body size

(elliptical body area) was statistically different among male *H. convergens* surviving diet

treatments ($F=5.3$, *df*=2,20, *p*=0.014), and between the control (1174 mm^2) and

sulfoxaflor (840 mm^2). In general, both male and female *H. convergens* were smaller in

the insecticide treatments versus the control, and this reduction could have significant

effects on fitness. Larger female *H. convergens* are more fecund (Vargas et al. 2012),

thus the trend of decreased size indicates sulfoxaflor may have a negative impact on lady

beetle reproduction. A reduction in body size could also negatively affect fecundity of

female *C. septempunctata* (Sundby 1968), but no size comparisons could be made as

there were no surviving adult females from the sulfoxaflor treatment.

Table 5: Effects of diet treatments on *H. convergens* and *C. septempunctata* cumulative
survival ratio (alive:total) at each larval stage and proportion female.

	Control	Flonicamid	Sulfoxaflor	Ratio Comparisons		
C. septempunctata				x^2	*df*	*P*
1st Instar	1.0a	1.0a	0.70b	10.0	2	0.007
2nd Instar	1.0a	1.0a	0.60b	14.9	2	<0.001
3rd Instar	1.0a	1.0a	0.60b	17.5	2	<0.001
4th Instar	0.95a	0.90a	0.40b	17.6	2	<0.001
Pupal	0.90a	0.90a	0.20b	30.3	2	<0.001
Proportion Female	0.44a	0.80b	0.0c	9.6	2	0.008
H. convergens						
1st Instar	1.0a	1.0a	0.90a	4.1	2	0.126
2nd Instar	0.95a	1.0a	0.90a	3.8	2	0.153
3rd Instar	0.95a	1.0a	0.70a	5.5	2	0.064
4th Instar	0.75a	0.90a	0.60a	5.5	2	0.064
Pupal	0.70a	0.80a	0.50b	7.0	2	0.030
Proportion Female	0.50a	0.50a	0.20a	5.7	2	0.060

Means in columns followed by the same letter are not statistically different (p <0.05) in a 2 x 2 χ^2 test.

Microcosm Study

The purpose of the microcosm study was to replicate field conditions in which Coccinellids compete with each other for prey when insecticides are introduced to the system causing acute toxic effects and additional strain as prey items decrease. Due to its delayed die-off effect, flonicamid is hypothesized to allow lady beetles in field environments to feed and survive longer than a pesticide that rapidly kills aphids.

Intraspecific interactions. For all replications and treatments, there were no aphids left on the plant after 3-4 days due to the combination of predation and/or insecticide effects. There were significant differences in the proportion of surviving *C. septempunctata* larvae for each 3-day interval evaluated among the treatments in the microcosm study (*p* > 0.002) (Table 6). The data clearly indicate that *C. septempunctata* survival is negatively affected by both insecticides, with sulfoxaflor having a more severe effect than flonicamid at day 6. Sulfoxaflor and likely cannibalism caused larval die-off in the *C. septempunctata* microcosms by day 9, where the control and flonicamid allowed for larval survival up to day 12.

Table 6: Proportion of surviving larvae (*H. convergens* and *C. septempunctata*) reared in microcosms exposed to three different treatments.

	Control	Flonicamid	Sulfoxaflor	Ratio Comparisons		
C. septempunctata				x^2	*df*	*P*
Day 3	1a	0.95a	0.75b	18.9	2	<.001
Day 6	0.75a	0.35b	0.05c	20.9	2	<.001
Day 9	0.55a	0.15b	0b	18.1	2	<.001

|						
Day 12	0.35a	0.05b	0b	12.4	2	0.002
H. convergens						
Day 3	0.95	0.80	0.80	2.4	2	0.308
Day 6	0.90a	0.50b	0.25b	17.4	2	<.001
Day 9	0.10	0.15	0.10	0.3	2	0.851
Day 12	0.05	0.05	0	1.03	2	0.596

Means in rows followed by the same letter are not statistically different (p <0.05) in a 2 x 2 χ^2 test.

$H. convergens$ survival was significantly impacted by both insecticides and/or cannibalism at day 6 ($p > 0.001$); the control and flonicamid allowed for larval survival through day 12, while sulfoxaflor treatments resulted in death of all individuals by day 6. Out of all replications, three *H. convergens* survived to adulthood from the control microcosms, while there were no surviving adults of either species from the two insecticide treatments. Trends for both species indicate that flonicamid microcosms allowed larvae to survive slightly longer than sulfoxaflor treated ones, likely because aphids are not rapidly killed when treated with flonicomid and therefore cannibalism was delayed.

Interspecific Interactions. Statistical analysis of the combined species microcosms (*H. convergens* + *C. septempunctata*) could not be preformed due to low replication, but trends indicate that both insecticides have negative effects on survival of both species as compared to the control (day 6-12) (Figure 10). No species survived to adulthood in this interspecific and limited diet competition test and species were not identified during larval stages to see who was surviving. The data suggests that the intensity of the interaction was greater than in the intraspecific tests, and given that there

were no surviving adults from any treatment, we can only speculate about the intraguild

predation dynamics. Mullins (2012) clearly demonstrated that larger *C. septempunctata*

larvae will readily eat *H. convergens* when aphid prey is limited and we suspect that these

dynamics occurred when the two predators were paired together.

Figure 10: Proportion of surviving larvae (2:2 *H. convergens* and *C. septempunctata*)
reared in microcosms exposed to three different treatments.
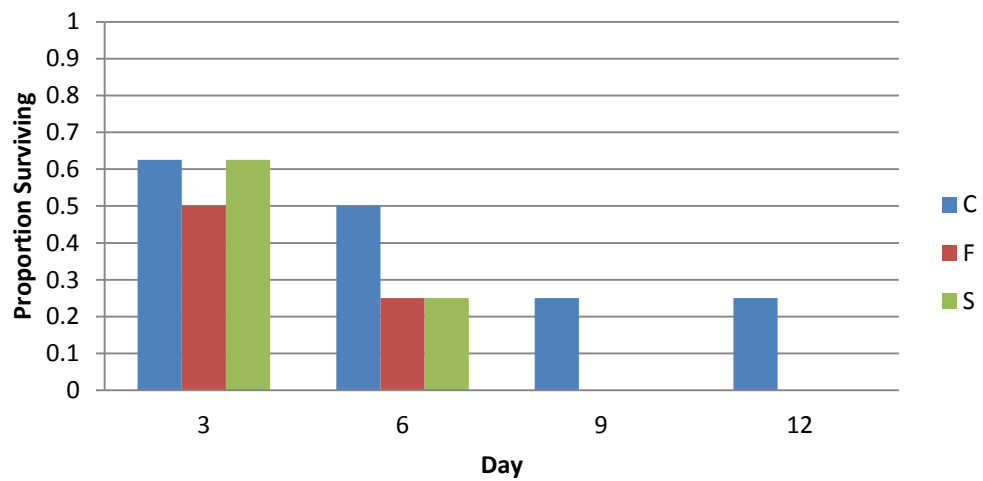
CHAPTER V

CONCLUSIONS

"Integrated control" programs (Stern et al., 1959) seek to utilize both biological and chemical control methods to reduce pest populations. Insecticides that have reduced negative effects on biocontrol agents, such as lady beetles, are vital to this concept. Synthetic pyrethroids, regularly used in winter canola in Oklahoma (Franke et al. 2009), are directly toxic to lady beetle and aphid species, so compounds with reduced acute toxicity would be desirable for management programs that are attempting to integrate biological and chemical control.

Lady beetle species immigrate and lay eggs in winter canola fields throughout the spring growing season. Both adults and newly hatched larvae may be exposed to insecticide treatments and forced to feed on altered or toxic food resources. Larval development time, adult body size, and male:female ratios from my trials indicate that flonicamid could be a valuable tool in fields where Coccinellids provide some level of biological control of aphids. Flonicamid allows for the possibility of longer-term lady beetle survivorship, where other pesticides such as synthetic pyrethroids do not.

In a preliminary field evaluation, three weeks were required for flonicamid to reduce

aphid populations to below the economic threshold (K. L. Giles, unpublished data). During this time, aphids (including newly born and those immigrating) are no longer able to feed and further crop damage does not occur, but aphids remain in the field and are fed on by lady beetles and other biological control agents. This may allow more lady beetles to continue development, which would support their overall populations in canola and surrounding landscape. Lady beetles reaching adulthood would be able to migrate into other fields and lay eggs, allowing system-wide population maintenance.

Conversely, this study indicates that sulfoxaflor treated aphids have a toxic effect on Coccinellids and may be a poor choice for use in similar fields. Consumption of sulfoxaflor treated aphids resulted in longer development times, which expose lady beetle larvae to other mortality effects, such as predation and further pesticide sprays. Sulfoxaflor treatments also resulted in reduced body size in adult lady beetles, which would likely decrease fecundity and ultimately reduce population size in the agricultural landscape. The fact that there were no surviving female *C. septempunctata* feeding on sulfoxaflor-treated aphids, and that male *H. convergens* were of smaller body sizes, indicates that sulfoxaflor-treated fields could be an ecological sink for these predator species.

This study indicates that fourth instar lady beetles are more susceptible to the negative effects caused by toxicity in their food sources. This may be because this instar requires more food than other instars to reach the pupal stage. It could also be because the lady beetle's body is chemically changing to transition into the pupal stage. All stages of larvae exist in the field at any given time, and migration heavily influences populations.

Lady beetles move in and out of interfacing wheat and pasture fields that surround canola fields in the landscape. They will readily migrate out of fields if no food is present. The use of narrow-spectrum insecticides that allow for survival and development to adulthood allows for lady beetles and other natural enemies to maintain populations throughout the system.

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VITA

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