THE EFFECT OF CHEMICAL CUES ON HOST

FINDING AND HOST ACCEPTANCE BEHAVIOR OF

DIAERETIELLA RAPAE (M'INTOSH)

(HYMENOPTERA: BRACONIDAE)

By

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Title of Study: RESPONSE OF *DIAERETIELLA RAPAE* (M'INTOSH)
(HYMENOPTERA: BRACONIDAE) TO HOST AND/OR HOST
PLANT SEMIOCHEMICALS
And   PARASITISM RATE OF *DIAERETIELLA RAPAE* (M'INTOSH)
(HYMENOPTERA: BRACONIDAE) IN AN EXPERIENCED VERSUS
A NOVEL ENVIRONMENT

Major Field: ENTOMOLOGY AND PLANT PATHOLOGY

Abstract:
        In winter wheat (*Triticum aestivum*) in Oklahoma, aphid pest suppression occurs through the natural activities of the parasitoid wasp *Lysiphlebus testaceipes* (Cresson). As winter canola (*Brassica napus*) has become increasingly grown as a rotation crop with wheat, it has not experienced the same kind of suppression for its aphid pests, namely the turnip aphid, *Lipaphis pseudobrassicae (Davis)* and the cabbage aphid, *Brevicoryne brassica* (L.). The principle cabbage aphid parasitoid, *Diaeretiella rapae* (M'Intosh) is one of the possible means of biological control. Its behavior in relation to volatiles present in both wheat and canola has been tested to determine the likelihood of this parasitoid to be used in an integrated pest management program. Although it does not learn chemicals in its immature stages inside the aphid host, this wasp is greatly influenced by volatiles it is exposed to upon emerging from its host. Depending upon its emergence environment, *D. rapae* can be an effective means of biological control with parasitism rates comparable to that of *L. testaceipes* on aphids in wheat. It is limited by its ability to learn and respond to chemical cues that are different from its emerging environment, most notably when the environment changes from canola to wheat and it experiences a very low rate of parasitism. Because canola is planted for only 1-2 years consecutively these small populations of *D. rapae* may struggle to reach high enough densities to be considered an effective means of aphid pest suppression in canola.

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CHAPTER I

INTRODUCTION

The primary crop produced in Oklahoma has historically been winter wheat (*Triticum aestivum* L.) with an average of 1.4 to 2 million hectares (3.5 to 5 million acres) planted annually over the last two decades (National Agricultural Statistics Service 2013). Additionally, these wheat acres are typically planted without rotation with other winter crops and the wheat is grown for forage, forage + grain or grain only (Epplin et al. 2000). This continuous wheat production system has supported increased weed and plant disease and arthropod pest problems in the fields (Bushong et al. 2012). Difficult to control annual grass weeds include cheat, *Bromus secalinus* L.; feral rye, *Secale cereale* L.; Italian ryegrass, *Lolium perenne* L. spp. *multiflorum* (Lam.) Husnot); jointed goatgrass, *Aegilops cylindrica* Host; and wild oat, *Avena fatua* L.. A 2009 survey showed that these five grassy weeds contribute economic loss from yield loss and loss due to discounted prices applied to harvested wheat that is contaminated with any of these weed seeds (Fast et al. 2009) in order to address these problems, a new winter crop was introduced; canola,

Brassica napus L. It has been shown to break disease cycles (Buntin et al. 2007), aid in weed management (Blackshaw et al. 2005), and improve net returns (DeVuyst et al. 2009, Bushong et al. 2012). Canola's market value is comparable to or higher than wheat and since its introduction acreage has increased (National Agricultural Statistics Service 2013).

While the history of wheat cultivation in Oklahoma has led to sophisticated IPM programs relying on natural enemies to control for insect pests (Giles et al. 2003, Royer et al. 2003, Elliott, et al. 2004), the same cannot yet be said for canola. Franke et al. (2009) surveyed Oklahoma canola producers about their production concerns growing canola. Growers listed insects as the second most important production issue and aphids as the most important insect pest of concern. With the primary pest complex of canola being aphids (Royer and Giles 2009), the development of comprehensive economic thresholds is important. While thresholds based on aphids alone are offered (Sanders et al. 2006), comprehensive thresholds including the impact of natural enemies are not available.

The Glance-n-Go aphid sampling system in wheat was developed from research conducted by Giles et al. (2003) which documented that a parasitoid wasp *Lysiphlebus testaceipes* (Cresson) needed to be considered when determining the need for control of greenbugs. The Glance n' Go system first considers abundance of *L. testaciepes* then greenbug density to determine the need to use an insecticide (Royer et al. 2003). *Lysiphlebus testaceipies* controls aphids by using the pest as a host to develop. In the process, the aphid loses reproductive capability and is eventually killed. The most commonly found aphids in wheat that can be used as hosts include the green bug,

Schizaphis graminum (Rondani) bird cherry-oat aphid, *Rhopalosiphum padi* (L.), and Russian wheat aphid, *Diuraphis noxia* (Kurdjumov) (Royer et al. 1997 and Silva et al. 2008). In wheat fields rotated with canola, the associated aphid pests also change and possibly the natural enemies that control them (Bullock 1992 and Wyland et al. 1996). Common aphid pests include the cabbage aphid, *Brevicoryne brassica*, green peach aphid, *Myzus persicae* (Sulzer) and the turnip aphid, *Lipaphis pseudobrassicae* (Davis) (Royer and Giles 2006). In northern canola growing regions *Diaeretiella rapae* (M'Intosh) is found utilizing the aphids present (Pike et al. 1999).

Diaeretiella rapae along with *L. testaceipes* are the most likely candidates to provide natural suppression of aphids in canola. To better understand the impact *D. rapae* will have in the field, its mechanisms for habitat and host location as well as its ability to utilize aphids from both wheat and canola needs to be determined. These studies are aimed at determining what cues present during *D. rapae*'s lifecycle are responsible for host habitat and/or host seeking behavior as well as its ability to parasitize aphids from a known habitat versus and unknown as wheat and canola are rotated in the same area.

Objectives

1. Determine the effect of the aphid mummy on recently emerged adult *Diaeretiella rapae* to host plant, host, and host/host plant combination.
2. Determine if there is a preference for odor source by *Diaeretiella rapae.*
3. Compare parasitism rate of *Diaeretiella rapae* in a novel plant and aphid environment and an experienced plant and aphid environment.

Explanation of Thesis Format

The chapters of this thesis describe the results of two experiments. The first is an analysis of host habitat and host finding behavior of *Diaeretiella rapae* in the presence or absence of chemicals associated with the host. The second is a breakdown and quantification of parasitism rate as well as incidence of superparasitism of either bird cherry-oat aphid in wheat or cabbage aphid in canola by *Diaeretiella rapae* originating from either experienced conditions (wheat to wheat and canola to canola) or novel conditions (wheat to canola and canola to wheat). Following the general introduction (Chapter I) is a detailed review of literature (Chapter II) including a list of references. Chapters III and IV are formatted according to the Entomological Society of America's standards with the associated references following each chapter.

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CHAPTER II

REVIEW OF LITERATURE

Winter Wheat

In total, roughly 2.4 ha (6 million acres) of wheat are planted annually and are used for grain, forage, and dual purpose forage and grain (Krenzer 1994). Winter wheat used solely for grain production is planted in mid-October and harvested in June and July. Wheat can be planted earlier in August and September if it is being used as a forage or dual-purpose crop respectively, but this causes a reduction in grain yield as a trade-off (Epplin et al. 2000;Hossain et al. 2003).   Grain-only accounted for 39% of the acres planted, forage-only for 22%, and 39% for dual-purpose.

In 2012 Oklahoma was the 4th largest producer of wheat harvested for grain with a yield of 42,172,175 quintals (154,800,000 bu), (NASS 2013). Oklahoma wheat production ranged from 19,070,105 to 48,764,983 quintals (70,000,000 to 198,900,000 bu) from 1994-2013. Highest winter wheat yields occurred in 1998 and lowest in 1996. These fluctuating yields can be attributed to varying climate (e.g. temperature and precipitation) during the growing season (NOAA 2013) and pests (diseases, insects and weeds).

Common aphid pests of wheat include the greenbug (*Schizaphis graminum* Rondani) (Hemiptera: Aphididae), Russian wheat aphid, *Diuraphis noxia* (Kurdjumov) (Hemiptera: Aphididae), corn leaf aphid, *Rhopalosiphum maidis*(L.) (Hemiptera: Aphididae), English grain aphid, *Sitobion avenae* (F.) (Hemiptera: Aphididae), rice root aphid, *Rhopalosiphum rufiabdominalis* (Sasaki) (Hemiptera: Aphididae), and bird cherry-oat aphid, (*Rhopalosiphum padi* (L.) (Hemiptera: Aphididae) (Blackman and Eastop 1984). Aphids are a significant pest of winter wheat (Giles et al. 2003) and can cause direct yield loss as well as act as a vector for barley yellow dwarf virus (Pike, 2007). There are several methods for aphid pest control practiced in wheat in Oklahoma. Traditional chemical controls in the form of insecticides were applied to 24% of the acres planted and equaled about 231,785 kg (511,000 lb) of insecticide (NASS, 2005). This is a reduction from 32% of planted acres from data reported in 2002 (NASS, 2002). Control measures specifically for bird cherry-oat aphid rely primarily on natural enemies and parasitoids in the spring when over-wintering populations begin to grow (Ahmad 2008; Summers and Godfrey 2009; Whitworth and Shoup 2012). Cultural methods include delayed planting dates and destruction of volunteer wheat (Whitworth and Ahmad 2008).

All of these methods are important not only to reduce yield loss, but also to reduce transmission of barley yellow dwarf virus (BYDV) from cereal aphid vectors such as the bird cherry-oat aphid (Pike 2007). BYDV, family Luteoviridae, symptoms include leaf tip discoloration (yellow or red) and stunting (Gray, 2010). These result in reduced seed yields (Banks et al. 1995). In transgenic crops the nymphal development of bird cherry-oat aphid is longer, providing more time for predators and parasitoids to locate the aphid (Jimenez-Martinez et al. 2004). On susceptible, untransformed wheat the nymphal

development time is much quicker and could facilitate the spread of the virus (Jimenez-Martinez et al. 2004). Wheat that has had the aphids that transmit BYDV removed has shown some ability to recover, but with reduced yield loss (Riedell et al. 2004).

Canola

Wheat used for grazing is commonly mixed with other plants, such as Italian ryegrass to lengthen the foraging season. Unfortunately these grasses can become detrimental and difficult to control if the wheat field will be harvested for grain in the future (Hossain 2004, Fast et al. 2009).

In 1997 a winter variety of canola (Wichita) was introduced to Oklahoma as a rotational crop with wheat in order to increase wheat yields and reduce pests (Boyles et al. 2009. In 2010 Oklahoma became the 2nd largest producer of canola and has maintained that place behind North Dakota. Area of acres planted increased by 130% from 24,000 ha (60,000 acres) in 2010 to (56,600 ha (140,000 acres) in 2012 and total production increased from 40,370,000 kg (89,000,000 lb) to 73,030,000 kg (161,000,000 lb) (NASS 2013).

Although canola was introduced to benefit wheat production through weed control, its market value is actually higher and allows growers to continue to profit while controlling for weeds and disease (DeVuyst et al 2009). Canola can be marketed for either edible oil or meal and offers an alternative to the grain market which has been the primary market for crop growers in Oklahoma (Boyles et al. 2009). There is also no additional machinery cost for wheat growers who rotate canola as small grains equipment can be used for both (Boyles et al. 2009 and Boyles et al. 2010).

In two surveys of canola growers, insect pests were listed as a major concern with aphids being the most economically important (Brown et al. 2008 and Franke et al. 2013). In Oklahoma, aphid pests of canola include the turnip aphid, *Lipaphis pseudobrassicae* (Davis) (Hemiptera: Aphididae), the green peach aphid, *Myzus persicae* (Sulzer) Hemiptera: Aphididae), the cabbage aphid *Brevicoryine brassicae* (L.) Hemiptera: Aphididae), the poplar petiole gall aphid, *Pemphigus populitransversus* Riley (Hemiptera: Aphididae), and the bean root aphid, *Smynthurodes betae* (Westwood) Hemiptera: Aphididae) (Eastop, 2000). Of these the turnip aphid, cabbage aphid, and green peach aphid are all recurring pests of canola (Royer and Giles 2013) as well as suitable hosts for the parasitoid wasp *Diaeretiella rapae* (M'Intosh Hymenoptera: Braconidae) (Pike et al. 1997 and Pike et al. 1999).

Current pest management strategies in canola rely on treatment of seed as well as approved insecticides like Warrior® and Capture® (Franke et al. 2013). Aside from pesticide use, IPM practices to promote natural enemies in canola have not been well developed or researched (Brown et al. 2008, Franke et al. 2013, and Royer and Giles 2013).

Bird Cherry-Oat Aphid; *Rhopalosiphum padi* (L.) (Hemiptera: Aphididae).

Bird cherry-oat aphids are known to feed on wheat as well as other cereal grain crops. In colder climates *R. padi's* lifecycle is holocyclic, but it is not obligatory (Macfadyen and Kriticos 2012). In Oklahoma these aphids are anholocyclic and typically stay in the parthenogenic phase year round. In the winter and early spring bird cherry-oat aphid can be found on winter wheat where it overwinters. In the spring, populations

migrate to other Poacae and bird cherry (hackberry) tree, *Prunus padus*, where it

continues asexual reproduction until the fall when females give birth to sexual males and

females (Dixon 1971 and Whitworth and Ahmad 2008).

Bird cherry-oat aphids are capable of utilizing both the leaves and fruiting portion

of the wheat plant. This allows the aphid to infest wheat from planting through harvest

(Honek et al. 2006). In addition to direct feeding damage to the plant, bird cherry-oat

aphids are a vector of barley yellow dwarf virus. This virus can cause stunting in the

plant, leaf necrosis, lack of heading and a red or yellow leaf discoloration which leads to

direct yield loss from reduced seed production (Comeau 1987 and Banks et al. 1995).

Additional damage from aphid and virus infestations is caused to the roots with infected

tillers having up to 40% root length reduction (Riedell et al. 2003). Plants infected with

barley yellow dwarf virus are less suitable than non-infected plants for aphids, likely due

to poorer nutritional quality caused by the presence of the virus (Jimenez-Marinez et al.

2004). The dislodgement of bird cherry-oat aphids by predators has been shown to spread

the virus across larger areas, but attacks by parasitoids result in little aphid movement and

reduce the area of viral infection (Smyrnioudis et al. 2001).

Cabbage Aphid: *Brevicoryne brassicae* (L.) (Hemiptera: Aphididae)

The cabbage aphid occurs throughout all temperate and warm temperate parts of

the world and has either a monoecious holocyclic lifecycle in colder regions or an

anholoyclic cycle where winter temperatures are more mild in places like Oklahoma

(Black man and Eastop 1984). Development time from nymph to adult varies from a

maximum of 36 days in the winter and a minimum of 8 days during the spring (Hafez 1961).

The cabbage aphid is a minor nuisance or a major pest depending upon when and where canola is being grown (Alford 2003). In colder climates low temperatures in the winter prevent the populations from increasing to large enough numbers to be a pest in the spring when canola flowers (Alford 2003). Because winter temperatures are warmer in Oklahoma, cabbage aphid can be a major pest in the fall when canola germinates as well as the spring during flowering (Gu et al. 2007). In southern regions of the U.S. cabbage aphids colonize canola fields in the fall and populations build through the spring (Buntin and Raymer 1994). There are no records of viruses transmitted by the cabbage aphid in canola in the U.S.; however there have been reports of transmission of beet western yellow virus by cabbage aphid in Australia (Gu et al. 2007 and Berlandier et al. 2010) making the status of cabbage aphids as a vector possible.

Diaeretiella rapae (M'Intosh) (Hymenoptera: Braconidae)

The solitary endoparasitoid, *Diaeretiella rapae* (M'Intosh), is in the family Braconidae and subfamily Aphidiinae. As a koinobiont, this wasp uses aphids as a host for larval development, pupation, and emerges as an adult from the aphid. *Diaeretiella rapae*'s ancestral range is in the Mediterranean region, Europe, the Middle East and Asia but has spread to North and South America.(Baer et al. 2004). This wasp is capable of using a variety of hosts and habitats including the ancestral host, cabbage aphid, and Russian wheat aphid. From these it has become able to use >60 aphid species (Pike 2007).

The lifecycle of *D. rapae* begins when a female wasp locates a suitable aphid host, and oviposits an egg inside the aphid. With higher host densities a female will only oviposit one egg per aphid host, but at lower host densities superparasitism is known to occur (Van Alphen and Visser 1990 and Kant et al. 2011). The larva develops inside the host consuming nutrients and killing the aphid within about 3 days until only the exoskeleton is left. A small hole is created by the wasp in the ventral side of the aphid exoskeleton and is glued to a substrate (Kant et al. 2011). The aphid exoskeleton is referred to as a 'mummy' and can be identified as parasitized without aid of a hand magnifier or microscope. The wasp then undergoes pupation and emerges through a circular opening created on the dorsal portion of the aphid's abdomen. After emerging the wasp will spend some time grooming by rubbing its antennae with its forelegs and exploring the surface of the mummy.

Post emergence *D. rapae* goes through the host searching process seen in most parasitoids: habitat location, host location, host acceptance, and host suitability. Each of these steps is influenced by chemicals from the plants and host (Read et al. 1970, Dicke and Sabelis 1988, Lewis and Martin 1990, Vet and Dicke 1992, and Jervis 2005). With parasitoids, habitat location and host location can be distinct or a combined process (Jervis, 2005). This means that *D. rapae* can be attracted to the plants without the host present or plants with feeding damage from the host. These behaviors are often described as attractant behaviors which direct the parasitoid's movement (Vet and Dicke 1992 and Jervis, 2005).

Once the parasitoid has been attracted to an area containing hosts, arrestant behaviors are exhibited such as patch foraging, host inspection and host acceptance. Host acceptance is influenced in part by the size and age of the host. Aphids that are larger (i.e. older) are chosen preferentially to smaller (younger) aphids (Kant et al. 2011 and Tazerouni et al. 2011). If host numbers are high compared to females searching for a host, *D. rapae* avoids previously parasitized hosts. If there are low numbers of aphids superparasitism increases, but in all cases only one adult wasp will emerge from a host (Kant et al. 2011). Host suitability is likely determined partially by oviposition. A study conducted by Kant et al. (2011) quantified ovipositor probes in unparasitized cabbage aphids from *D. rapae* and also the number of larvae present in superparasitized aphids. When attacking unparasitized aphids there was, on average, 7.2±0.6 probes for a single host aphid, but after hosts were dissected those that were superparasitized only contained 2-5 wasp larvae. This gives evidence that counting the number of probes is not representative of the number of larvae present. As the wasp continues to search and oviposit, it demonstrates learning through measurable experience. Older wasps react differently to new hosts, parasitized hosts, and patch marking chemicals than younger, inexperienced wasps (Hérard et al. 1988, Van Alphen and Visser 1990, Vaughn et al. 1996, and Jervis 2007).

Of special interest is habitat and host location; *D. rapae* has shown a preference for cruciferous plants (Cruciferacae) and the mustard oils these plants secrete when damaged (Read et al. 1970 and Reed et al. 1994, Vaughn et al. 1996). They are also known to parasitize aphid hosts on non-cruciferous plants like *Myzus persicae* on the potato plant (Reed et al. 1994) and *Rhopalosiphum padi* in wheat (Pike et al. 1997).

Role of Semiochemicals

There are numerous cues that parasitoids can use to locate their host. They include visual, sound, and chemical cues. Visual cues are more likely to be used by parasitoids whose host is found within fruits; dipterous parasitoids commonly use sound to locate hosts like crickets; and the predominant cue in Hymenoptera is chemical (Jervis 2005), referred to as allelochemicals (Dicke and Sabelis 1988). The specific group of chemicals likely being detected by *D. rapae* is kairomones (Jervis 2005) as defined by Dicke and Sabelis (1988).

The relationship between plants, herbivores, and natural enemies is mediated by the chemicals associated with each (Price et al. 1980). In agricultural environments during a particular growing season, the system is typically a monoculture of early-succession plants (Price et al. 1980) that are more palatable and susceptible to herbivory but overcome these characteristic by rapid growth (Cates and Orians 1975 and Price 1991). A consequence of these high rates of herbivory is the release of chemicals by the plants (Price et al. 1980, Du et al. 1996, and Rose et al. 1996) that some aphid parasitoids, including *D. rapae*, can recognize (Wickremasinghe and van Emden 1992 and Reed et al. 1994). The damage-induced release of chemicals through the interaction between the herbivore and the plant has been shown to be more attractive to parasitoids than plants that are artificially damaged (Loke and Ashley 1984, Drost et al. 1986, Hérard et al. 1988, Turlings et al. 1988, and Du et al. 1998).

In addition to plant-stress related chemicals, those semiochemicals associated with the pest can also be used by parasitoid wasps (Loke and Ashley 1984, Lewis and

Martin 1990). These cues are important once the parasitoid has entered the host's habitat and, in addition to location, may be used to determine the suitability of a host and whether it is already parasitized (Vinson and Guillot 1972, Lewis and Martin 1990, and Nufio and Papaj 2001).

Hopkin's Host Selection Principle

Originally the chemical cues of some phytophagous and parasitic insects were thought to be learned in the larval stage, then transferred to the adult (Hopkins 1916). This implied that insects capable of developing in multiple environments will prefer the environment they actually developed in. This theory (Hopkin's host selection principle) has some research support with parasitoids (Sheehan and Shelton 1989 and Geervliet et al. 1996).

There are some basic assumptions about the wasps' innate preference in relation to experience that may weaken these conclusions. A wasp that is defined as 'naïve' or 'inexperienced' is described as having been restricted from live hosts and host plants, but wasexposed to the remains of the host as an adult (Sheehan and Shelton 1989, Geervliet et al. 1996). This means that exposure to the dead host and any semiochemicals associated with it are being overlooked as possible sources of learning. Studies refuting Hopkin's principle focus on this critical time and attempt to differentiate between adult learning and innate response by various methods including removal of parasitoid pupae from the host (Monteith 1962, Jaenike 1982, Caubet et al. 1992, van Emden et al. 1996, and Storeck et al. 2000).

Evidence for this learned vs. innate response to the host plant, host, and damaged host plant has been collected through a variety of methods including various studies using olfactometers and through electroantennogram analysis (Read et al. 1970, Reed et al. 1994, and Vaughn et al. 1996). These experiments studied behaviors within general populations (Read et al. 1970), male and female wasps individually (Vaughn et al. 1996), and inexperienced and experienced female wasps (Wickremasinghe and Van Emden 1992 and Reed et al. 1994). An 'experienced' female wasp can refer to several treatments including exposure to the host and environment or to ovipositional experience (Wickremasinghe and van Emden 1992 and Reed et al. 1994). Because of the variation of the term 'experience' and its inability to describe a wasp removed from the mummy, the term 'excised' as described in van Emden et al. (1996) is more appropriately used in reference to wasps removed prematurely from the mummy. Experiments with excised wasps have shown that many of the preferences attributed to an innate response have been misplaced and the cues are likely picked up during adult conditioning (van Emden et al. 1996 and Storeck et al. 2000).

Parasitism Rate

As the crops and pests change from year to year, *D. rapae* must be able to switch and utilize the available host to persist in the environment. Although local extinctions may occur, the metapopulation must remain if *D. rapae* is to be considered a necessary component of aphid control (Weisser 2000).

While there is some difference between results for rate of parasitism in field versus laboratory trials, basic laboratory experiments are an important foundation for

further testing (Lopez et al. 1990). This greatly reduces variability that is inherent in the field related to climate and weather and pest population behaviors (Lopez et al. 1990).

Both cabbage and bird cherry-oat aphids are similar in size with cabbage alatae ranging in size from 1.6 to 2.8 mm and apterae from 1.6 to 2.6 mm and bird cherry-oat aphid apterae and alatae ranging from 1.2-2.4mm in length (Blackman and Eastop 1984). While cabbage aphids tend to clump together along stalks of canola, especially at the plant nodes (Way and Cammell 1970), bird cherry-oat aphids are more aggregated along the wheat stalks (Quiroz et al. 1997).

Because there is a host density effect on *D. rapae* (Fathipour et al. 2006), steps to limit its impact on the quantification of parasitism can be taken in the form of manual aphid scattering (Lopez et al. 1990), aphid placement on leaf sections/disks (Rodrigues and Bueno 2001), and use of relatively equal numbers of aphids (Rodrigues and Bueno 2001 and Jones et al. 2003). The first two methods are unsuitable when comparing across environments because the wasps' normal searching behavior is being impacted. Instead, whole plants that are similar in overall surface area will be infested with equal numbers of aphids. This allows for any effects of plant morphology and aphid distribution to be minimized and make the host encounter rate similar between the two environments (Gingras et al. 2008).

In addition to pest density, intraspecific densities also affect parasitism with decreased parasitism observed as wasp density increases relative to a stable pest population (Fathipour et al. 2006). In the case of *D. rapae*, highest parasitism for an individual female occurs when there is no interference by other females (Fathipour et al.

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2006). The age of the female also affects attack rates: a study by Blande et al. (2004) showed that newly emerged wasps attacked at a lower rate than those that were 1-5 days old, within which there was no significant difference. Ovipositional experience also affects the choices and searching habits of parasitoids, so using inexperienced wasps that are a day old decreases preference variation due to age and past experience while maintaining the higher level of attack (Van Alphen and Visser 1990, Vaughn et al. 1996, Blande et al. 2004, and Jervis 2007).

Future Use as Biological Control

In Oklahoma, the dominant aphid parasitoid in the landscape is *Lysiphlebus testaceipes* and it is capable of maintaining aphid populations in wheat below economic thresholds (Elliott et al. 2014). Although there is speculation that this parasitoid (as well as *Diaeretiella rapae*) will move into canola, it is not assured that a similar level of control will be present. Competition between *L. testaceipes* and *D. rapae* in both wheat and canola is also possible because of host overlap on cereal aphids such as the green bug, green peach aphid, and bird cherry-oat aphid (Pike et al. 1999) but no record of *L. testaceipes* utilizing cabbage aphids as a host has been found (Desneux et al. 2006).

In other areas where canola has a longer history of being planted, there is a greater presence of *D. rapae* in canola in: Tennessee (Boyd et al. 1994), France (Desneux et al. 2006), and Iran (Lotfalizadeh 2002). There is evidence for partial aphid suppression in these areas and the suggestion of using *D. rapae* in an IPM program (Desneux et al. 2006).

Basic information about this parasitoid and its relationship with the common canola pest in Oklahoma will provide a foundation for continued research into its possible use as a biological control agent. As canola acreage increases across the landscape, the value of incorporating *D. rapae* into an IPM program needs to be ascertained.

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CHAPTER III

RESPONSE OF *DIAERETIELLA RAPAE* (M'INTOSH) (HYMENOPTERA: BRACONIDAE)

TO HOST AND/OR HOST PLANT SEMIOCHEMICALS

Introduction

For parasitoid wasps chemical cues are important for locating suitable hosts for

oviposition (Jervis 2005). Much work has been done to illustrate the ability of plants and

hosts to act as attractants to parasitoids of aphids (Reed et al. 1995, van Emden et al.

1996, Vaughn et al. 1996, and Storeck et al. 2000). In generalist aphid parasitoids, the

cues must be learned at some point during the wasp's development (Takemoto et al.

2011).

Following its introduction, Hopkin's host selection principle has been attributed to larval

development and exposure through the host (Geervliet et al. 1996). More recently

research involving the removal of wasp pupae from the dead host, or 'mummy', has

demonstrated that there is no response to chemical cues from these excised wasps. It was

concluded that adult exposure to the mummy (either as the wasp chews through or after

emergence) is the time when the chemical cues are learned (van Emden et al. 1996 and

Storeck et al. 2000). The parasitoid *Diaeretiella rapae* (M'Intosh) (Hymenoptera:

Braconidae), is a primary parasitoid of aphid pests found on Cruciferous plants but is also

known to use a variety of aphids on plants from different families (Pike et al. 1999). In Oklahoma it has shown potential to control cabbage aphid, *Brevicoryne brassica* (L.) (Hemiptera: Aphididae) and turnip aphid; *Lipaphis pseudobrassicae* (Davis) (Hymenoptera: Aphididae) associated with canola (*Brassica napus*). If *D. rapae* is to be an important aphid parasitoid in both canola and the primary winter crop, wheat (*Triticum aestivum*), it will need to easily switch between hosts. Canola is used in rotation with wheat and is typically only planted for a single growing season before wheat is planted again. This makes it transient in the landscape and populations of *D. rapae* would be forced to use a wheat monoculture instead the following year if wild mustards are not present in the surrounding landscape.

This study aims to evaluate the response of *D. rapae* from both wheat and canola under control conditions and those excised from mummies. Response to plants, hosts, and a combination of plant and host will be used as odor sources for *D. rapae.* Additionally, sources of odor will be compared to determine which is the best attractant.

Materials and Methods

Colony Maintenance. Bird cherry-oat aphids (BCOA) and cabbage aphids (CA) were taken from laboratory colonies maintained at Oklahoma State University (Stillwater). Aphids were transferred to fine mesh, single-walled cages onto approximately three week old plants. Bird cherry-oat aphids were reared on susceptible wheat (c.v. 'Jagger') and cabbage aphids on canola (c.v. 'Wichita'). Cages were kept at 22±1°C and 16:8 L:D. New wheat and canola plants were introduced as needed to maintain the aphid colonies.

Diaeretiella rapae were collected from laboratory stock maintained at the USDA-ARS facility in Stillwater, OK. Stock was reared on turnip aphids in canola and collected as both mummies and adults and a mix of both stages was introduced into either wheat infested with bird cherry-oat aphids or canola infested with cabbage aphids. Cages were constructed with double-walled, fine mesh (Jones et al. 2003) and kept at ambient lab temperatures of 22±1°C and L:D of 16:8. Adult wasps were collected and moved between each cage for each generation to maintain genetic diversity of the colonies.

Mummies of *D. rapae* were collected from both wheat and canola and stored individually in 2.9 x 2.74 cm (1.15x1.08 inch) round Polyvials™ under the same conditions as aphid colonies. Mummies were either stored until adult wasp emergence (control) or excised as described in van Emden et al. (1996). Wasps were used no more than 24h after emergence.
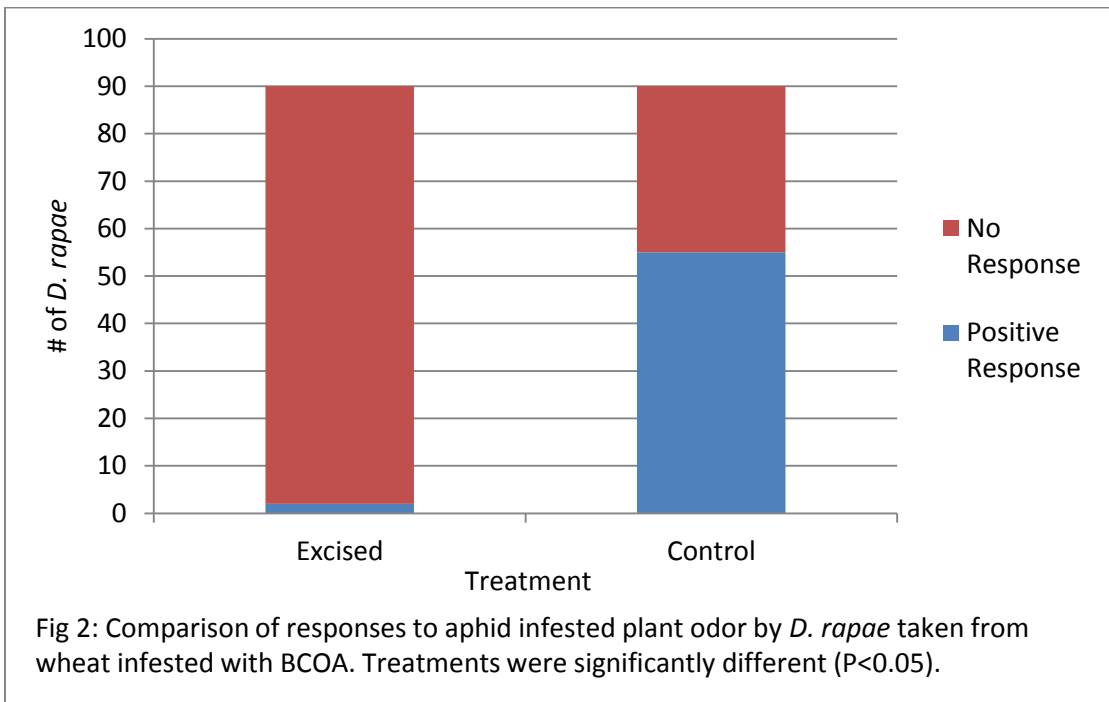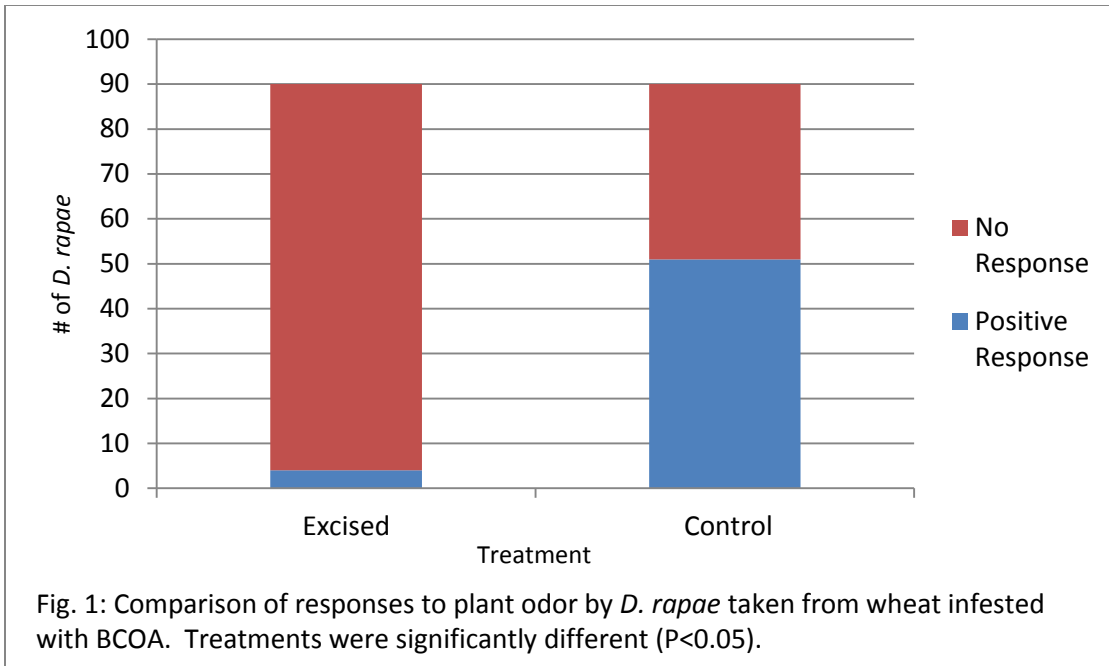
Experimental Methods. A two-armed olfactometer was provided as an arena for observation of wasp behavior. The olfactometer was placed directly under a single light source and air was suctioned from the two arms through the body of the tube towards the entrance of the apparatus. Three treatments were used to test the response of both control (n=90) and excised (n=90) wasps to different semiochemical sources. The first treatment used undamaged wheat and undamaged canola as a source. The second used bird cherry-oat aphid (n≈50) and cabbage aphid (n≈50) and the third used wheat infested with n≈50 bird cherry-oat aphids and canola infested with n≈50 cabbage aphids. Each treatment was conducted twice with either *D. rapae* collected from laboratory colonies of wheat infested with bird cherry-oat aphids or *D. rapae* collected from laboratory colonies of canola infested with cabbage aphids.
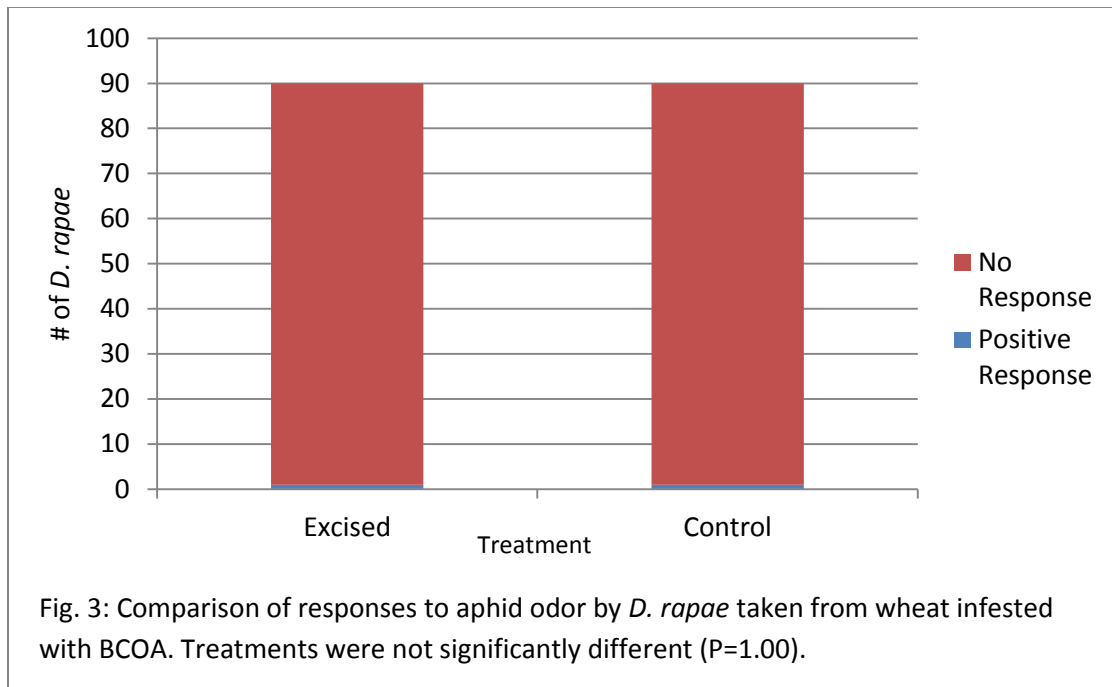
For each trial a single wasp was introduced into the opening of the olfactometer and observed for movement along the main tube and into the branches. Outcomes were recorded as follows: no movement into the tube or returning to the entry after two minutes was 'no response', movement into the main tube but not into either of the arms for ≥30s was 'no preference', and movement into either of the arms for ≥30s was recorded according to the source of the chemical in that branch. Each trial ran for a maximum of seven minutes if other criterion were not met.

All statistical tests were conducted using SAS™ 9.3 (SAS Institute 2014) with a significance level α=0.05. Fisher's exact test (PROC FREQ) was used to determine the difference in control versus excised wasps' responses to the three treatments for *D. rapae* coming from wheat and those coming from canola. Differences in behavior between *D. rapae* from canola versus those from wheat were determined using chi-square (PROC FREQ). Populations from both wheat and canola were combined and differences between treatments were determined using chi-square (PROC FREQ).

Results

The control group of *D. rapae* taken from wheat was more responsive to the odors of undamaged wheat and canola than those *D. rapae* excised from the mummy (Fig. 1: df = 1, $P < 0.001$). Similarly, *D. rapae* in the control group exposed to a combination of wheat/BCOA and canola/CA odors responded more than those that were excised (Fig. 2: df=1, p<0.0001). Those *D. rapae* taken from wheat and exposed to odors from BCOA and CA showed little response in either the control or excised group (Fig. 3: df = 1, $P = 1.000$).

Fig. 1: Comparison of responses to plant odor by *D. rapae* taken from wheat infested with BCOA. Treatments were significantly different (P<0.05).

Fig 2: Comparison of responses to aphid infested plant odor by *D. rapae* taken from wheat infested with BCOA. Treatments were significantly different (P<0.05).

Fig. 3: Comparison of responses to aphid odor by *D. rapae* taken from wheat infested with BCOA. Treatments were not significantly different (P=1.00).

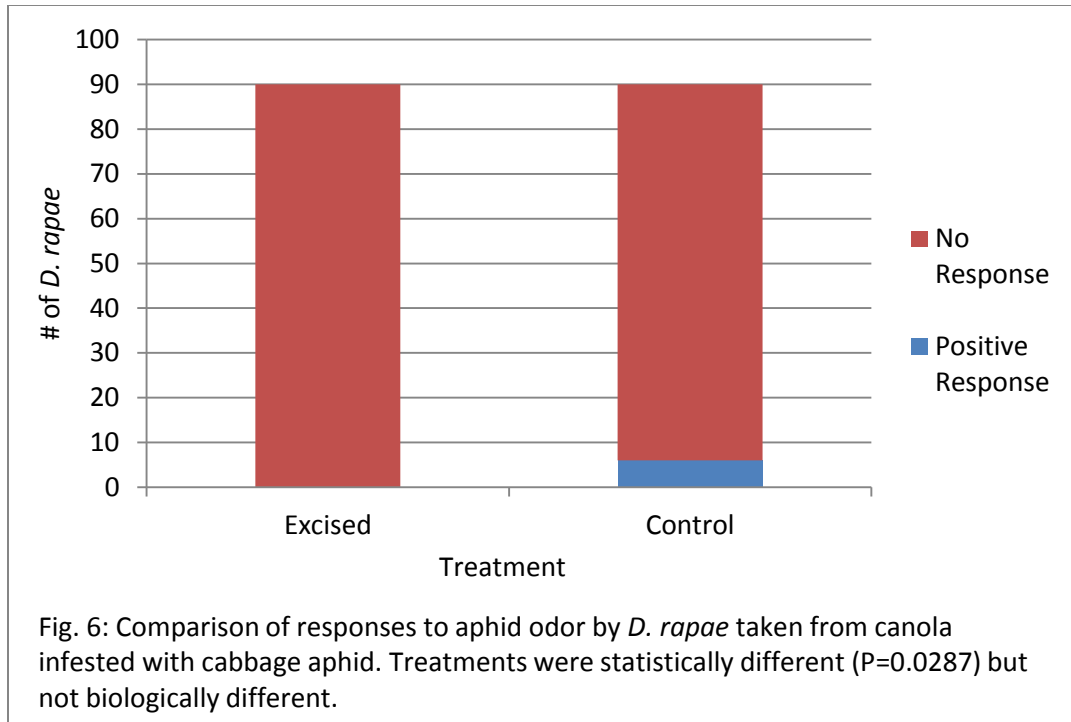Populations of *D. rapae* taken from canola exhibited similar patterns with the control wasps having a stronger response to the odor of undamaged plants compared to excised wasps (Fig. 4: df = 1, $P < 0.0001$). In the treatment where a combination of plant and aphids was used as an odor source *D. rapae* that had not been excised were more responsive (Fig. 5: df = 1, $P < 0.0001$). When only BCOA and CA were provided as an odor source neither the control or excised *D. rapae* groups showed a response to the odors but there was a significant difference in the populations (Fig. 6: df = 1, $P = 0.0287$) but it was not biologically significant.

The different odor sources provided varying results as attractants (Fig. 7: $\chi^2 =$ 183.82, df = 2, $P < 0.0001$). When the two aphid species were used as a source there was little attraction (3.9% positive response). Both the undamaged plant treatment and combination of plants and aphids treatment have a higher percentage of wasps attracted

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Fig. 4: Comparison of responses to plant odor by *D. rapae* taken from canola infested with cabbage aphid. Treatments were significantly different (P<0.05).

Fig. 5: Comparison of responses to aphid infested plant odor by *D. rapae* taken from canola infested with cabbage aphid. Treatments were significantly different (P<0.05).

Fig. 6: Comparison of responses to aphid odor by *D. rapae* taken from canola infested with cabbage aphid. Treatments were statistically different (P=0.0287) but not biologically different.

(58.3 and 70.6% respectively). A comparison of plant odor source to aphid showed that plants were a greater attractant ($\chi^2 = 124.48$, df = 1, $P < 0.0001$) and the combination of plant and aphid source was also a better attractant than aphids alone ($\chi^2 = 171.18$. df = 1, $P < 0.0001$). Additionally, when the odor sources of undamaged plants was compared to the combination of plants and aphids the combination was a better attractant ($\chi^2 = 5.87$, df = 1, $P = 0.0154$).

When comparing *D. rapae* from wheat to those from canola in treatments where there was a greater positive response to the odor, the populations taken from wheat and canola had similar levels of attraction to undamaged plant odors (Fig. 8: $\chi^2 = 0.6501$, df = 1, $P = 0.7625$) but wasps taken from canola had a higher incidence of attraction to infested wheat and canola than wasps taken from wheat ($\chi^2 = 7.7284$, df =1, $P = 0.0085$).
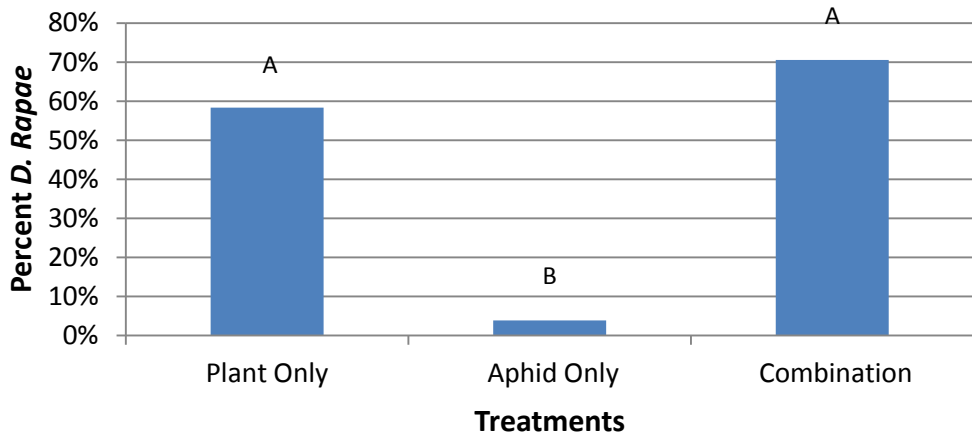
Fig. 7 Percentage of *D. rapae* responding to undamaged wheat and canola (Plant Only), BCOA and CA (Aphid Only), or wheat infested with BCOA and canola infested with CA (Combination). Columns with the same letter are not significantly different ($P < 0.05$).

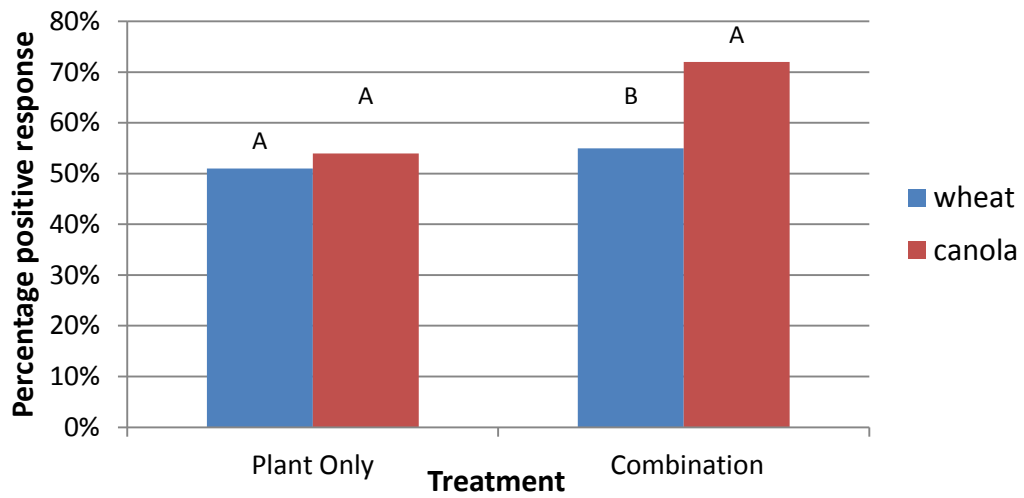Fig. 8 Percentage of *D. rapae* responding to undamaged wheat and canola (Plant Only) or wheat infested with BCOA and canola infested with CA (Combination). Columns with the same letter within each treatment are not significantly different ($P < 0.05$).

Discussion

By depriving *D. rapae* from contact with the chemicals associated with their mummies it was possible to determine that this contact is important for the wasp to be

able to recognize and locate odors associated with habitat and host location. Similar results were obtained by van Emden et al. (1996) for *Aphidius rhopalosiphi* (De Stef.) and Storeck et al. (2000) for *Aphidius colemani*. Our findings provide further support for the idea that the chemical legacy (Corbet 1985), in this case the contact with the mummy's skin by the adult, is responsible for the subsequent host habitat and host finding behaviors.

It is likely that, should the excised *D. rapae* be exposed directly to a mummy taken from either environment that wasp will then exhibit positive taxis towards those odors. A 'learning curve' is often associated with generalist predators and parasitoids and after repeated exposure the response to those volatiles will increase (Storeck et al. 2000).

Our finding that aphids feeding on plants are the most attractive odor source to *D. rapae* are similar to those found by other authors specifically for *D. rapae* (Read et al. 1970, Reed et al. 1995, and Vaughn et al. 1996) as well as other aphid parasitoids (Du et al. 1996, Geervliet et al. 1996, and Du et al. 1998) These results support the ideas presented by Vet and Dicke (1992) that, although the odors from the host are the most reliable, they are also difficult to detect. Chemical's produced by the plant in reaction to feeding are a little less reliable, but are much easier to detect and give the parasitoid a better chance of finding its host in the larger landscape.

This does not explain why, when *D. rapae* is taken from wheat with bird cherry-oat aphid as a host, it is less responsive to odors from infested wheat and canola than those taken from canola. This could be attributed to the ability of the wasp to respond to both odors given their prior experience. When Vaughn et al. (1996) used an

electroantennogram to test the reaction of *D. rapae* to volatiles associated with wheat and cruciferous plants the response to the latter was much stronger. The ability of *D. rapae* to recognize and then respond when already exposed to canola would likely be better than those with no prior experience (Sheehan and Shelton 1989).

Within the winter agricultural landscape of Oklahoma these findings have several implications for the ability of *D. rapae* to recognize and locate isolated patches of canola within the larger wheat landscape. With evidence that *D. rapae* operates within fields (Vaughn and Antolin 1998) initial chemical learning is important for subsequent habitat and host location. Although their presence in wheat is very minimal (French et al. 2001 and Elliott et al. 2014), those *D. rapae* that emerge in wheat are likely to stay within that field instead of moving into canola. This may explain why *D. rapae* has been slow to move into canola fields especially since these are transient and are usually only planted for a year before returning to wheat the following year. Additionally, effects of temperature on *D. rapae* also reduced parasitism. Bernal and Gonzáles (1997) determined that *D. rapae* kept at 10°C went through a pre-oviposition period of 0.95 ± 0.26 days but at higher temperatures (21.1 and 26.7°C) successful parasitism occurred within the first day of emergence. Because regional populations have exhibited different development times under the same temperature (Campbell et al. 1974), local populations in Oklahoma may not have the same response to lower temperatures.

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CHAPTER IV

PARASITISM RATE OF *DIAERETIELLA RAPAE* (M'INTOSH) (HYMENOPTERA:

BRACONIDAE) IN AN EXPERIENCED VERSUS A NOVEL ENVIRONMENT

Introduction

In generalist aphid parasitoids the rate at which they oviposit and parasitize is different for the various aphid species they are able to use as a host (Godfray 1994). This can be influenced by several factors including attack rate, host density effects, and effects of the environment on the parasitoid (Jones et al. 2003, Fathipour et al. 2006, and Gingras et al. 2008). In an agricultural environment where host plant and host species changes throughout the year the wasp must be able to switch hosts or use resource patches to persist (Taylor et al. 1993).

The aphid parasitoid *Diaeretiella rapae* (M'Intosh) (Hymenoptera: Braconidae) is the principle parasitoid of aphids on Cruciferous crops but also has hosts on cereal grain crops (Pike et al. 1999). In Oklahoma this parasitoid has been present in wheat, but in much lower numbers than the primary winter wheat parasitoid *Lysiphlebus testaceipes* (Cresson) (Hymenoptera: Braconidae). With the introduction and increase in acreage of winter canola (*Brassica napus*) over the past ten years the populations of *D. rapae* are

likely to change since *L. testaceipes* does not use either primary aphid pest of canola as a host (Elliott et al. 2014).

Although the most accurate measure of actual parasitism rate involves detection of eggs inside the host, these are very small and difficult to find (Hofsvang and Hågvar 1983). Allowing the wasps to fully develop is an easy way to get an idea of successful parasitism, but this excludes those eggs laid in hosts that die before the wasp has fully developed. Instead, dissections of live aphid hosts between oviposition and mummy formation is used to get an estimation of parasitism.

These experiments were conducted to determine how parasitism rates of aphids from wheat and canola by *D. rapae* are affected by the environment from which the wasp originated.

Materials and Methods

Colony Maintenance. Bird cherry-oat aphids, *Rhopalosiphum padi* (L.), (BCOA) and cabbage aphids, *Brevicoryne brassica* (CA) were collected from established laboratory colonies maintained at Oklahoma State University (Stillwater). Aphids were transferred to fine mesh, single-walled cages onto approximately three week old plants. Bird cherry-oat aphids were reared on susceptible wheat (c.v.'Jagger') and cabbage aphids on canola (c.v. 'Wichita'). Cages were kept at $22\pm1°C$ and 16:8 L/D. New wheat and canola plants were introduced as needed to maintain the colonies.

Diaeretiella rapae were collected from laboratory stock maintained at the USDA-ARS facility in Stillwater, OK. Stock was reared on *L. ersimi* in canola and collected as both mummies and adults and a mix of both stages was introduced into either wheat

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infested with BCOA or canola infested with CA. Cages were constructed with double-walled, fine mesh and kept at ambient lab temperatures of about 22±1°C and L/D of 16:8. Adult wasps were collected and mixed between colonies each generation to maintain genetic diversity of the colonies.
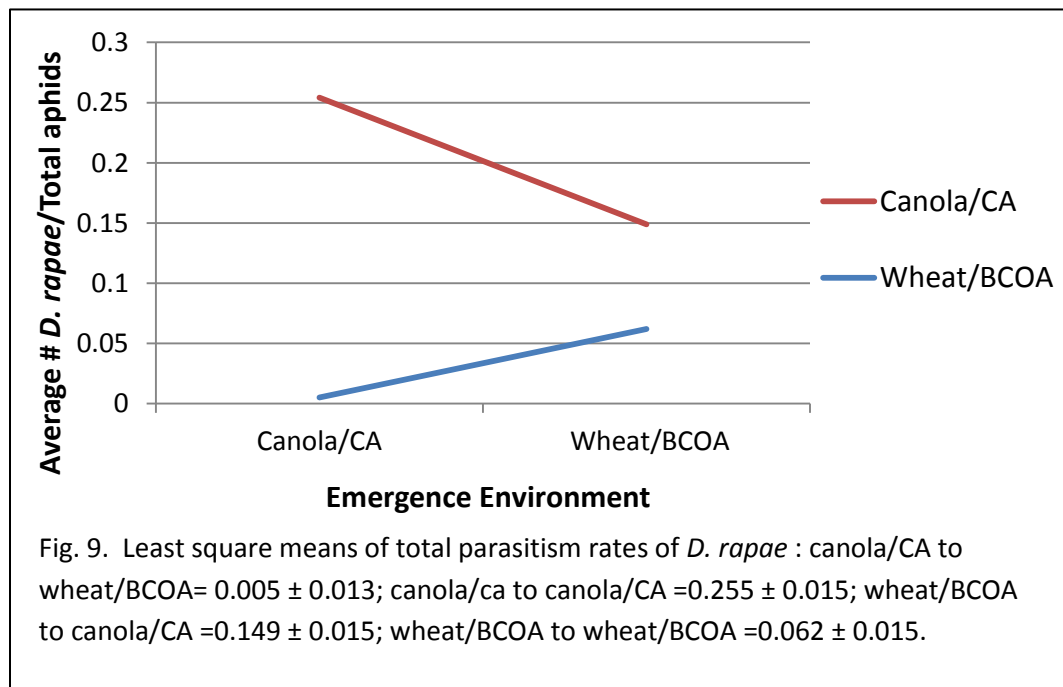
Experimental Methods. Individual treatments were constructed using a container for the plant roots and a clear, acrylic tube. Each tube has openings along the side and one on the top covered with fine mesh to allow air-flow. Either two wheat plants at 15 cm (6 inches) in height or two canola plants 10-15 cm (4-6 inches) tall or at the four-leaf stage were dug up by the roots and wrapped in cotton balls. Plants were placed into containers and water was added so cotton was damp. About 50 aphids were transferred from infested plants into containers using a fine brush and were allowed to settle for 24h prior to introduction of a single female *D. rapae*. Wasp mummies were taken from either wheat infested with BCOA or canola infested with CA (emergence environments) and stored individually until emergence. Once the adult emerged from the mummy it was then introduced into either wheat infested with BCOA or canola infested with CA (introduced environment). Wasps were removed after 24hrs and aphids were left for another two days before being collected.

Aphids that were dead after that time were collected separately from live aphids. All aphids were dissected and described as either: un-parasitized, parasitized, or superparasitized. Further categories of complete and incomplete parasitism were used to distinguish between parasitized aphids that were alive at time of collection or those that were dead, respectively.
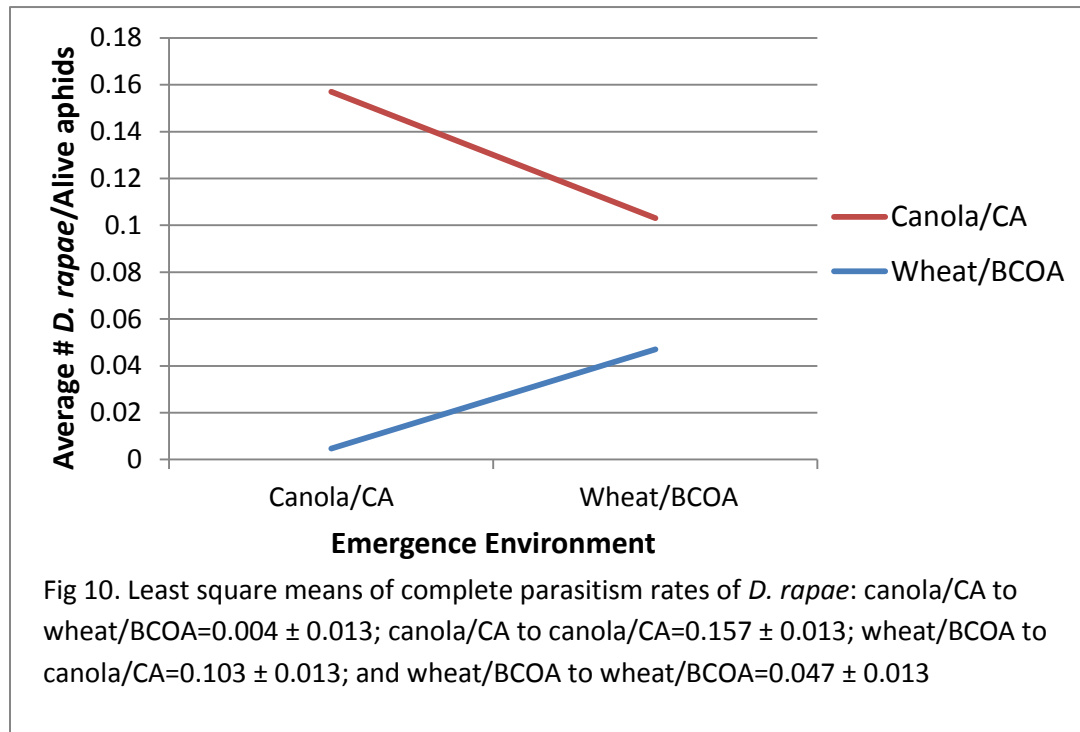
The effect of the emergence environment of the wasp, the environment the wasp was introduced to, and the interaction between the two were analyzed and the mean parasitism rates were fit to a linear model for total, complete, and incomplete parasitism rate. (PROC MIXED SAS Institute 2001).

Results

Total Parasitism. (Fig. 9). Total parasitism for each set of conditions was calculated by obtaining the average number of larvae per total aphids (both alive and dead at time of collection. There is a significant interaction between both emergence as well as introduced environment ($F = 13.24$, df = 36, $P = 0.0009$). This means that, regardless of emergence environment, being introduced into canola results in higher parasitism rates ($F = 65.87$, df = 36, $P < 0.0001$) and being introduced into wheat results in significantly lower parasitism rates ($F = 8.82$, df = 36, $P = 0.0053$).

Fig. 9. Least square means of total parasitism rates of *D. rapae* : canola/CA to wheat/BCOA= 0.005 ± 0.013; canola/ca to canola/CA =0.255 ± 0.015; wheat/BCOA to canola/CA =0.149 ± 0.015; wheat/BCOA to wheat/BCOA =0.062 ± 0.015.

Complete Parasitism. (Fig. 10) The complete parasitism rate, calculated from total number of larvae present in living hosts, had a significant interaction between the emergence and introduced environments ($F = 13.24$, df = 36, $P = 0.0009$). This interaction results in higher parasitism rates for *D. rapae* that are introduced into

Fig 10. Least square means of complete parasitism rates of *D. rapae*: canola/CA to wheat/BCOA=0.004 ± 0.013; canola/CA to canola/CA=0.157 ± 0.013; wheat/BCOA to canola/CA=0.103 ± 0.013; and wheat/BCOA to wheat/BCOA=0.047 ± 0.013

Incomplete Parasitism. (Fig. 11). Incomplete parasitism is the number of larvae present out of total dead aphids. There is a significant interaction between the emergence and introduction environments ($F = 9.23$, df = 36, $P = 0.0044$). The incomplete parasitism rate was higher for *D. rapae* that were introduced into canola ($F = 40.01$, df = 36, $P < 0.0001$) than for *D. rapae* introduced into wheat ($F = 4.11$, df = 36, $P = 0.05$) regardless of their emergence environment.

Fig. 11. Least square mean incomplete parasitism rate for each combination of conditions: canola/CA to wheat/BCOA≈0.00 ± 0.011; canola/CA to canola/CA=0.097 ± 0.011; wheat/BCOA to canola/CA= 0.045 ± 0.011; and wheat/BCOA to wheat/BCOA=0.014 ± 0.011.
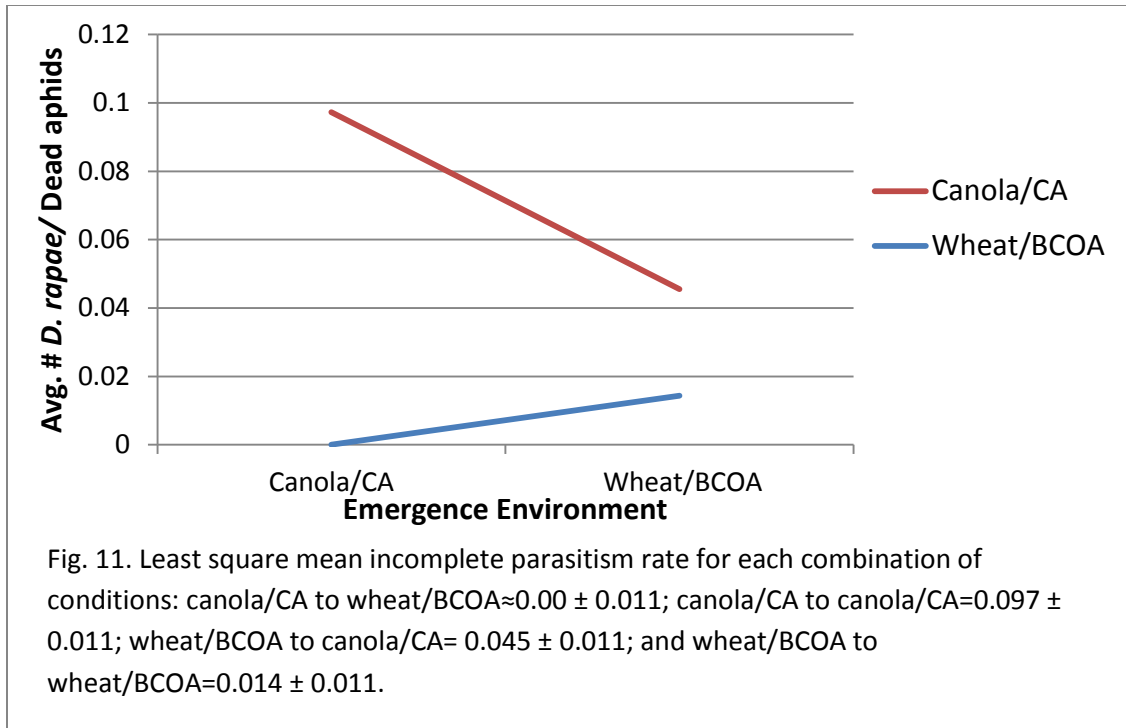
Discussion

For all three types of parasitism the complex interaction of emergence environment and introduced environment affected the average parasitism rate. Highest average rates of parasitism were highest for *D. rapae* coming from canola and introduced into it.

The lowest rates were from wasps emerging from canola and introduced into wheat. Although it could be argued that this is caused by a lag between the parasitoid and host recognition, the same low rates were not seen when *D. rapae* was taken from wheat and introduced into canola.

By dissecting aphids after such a short time period from initial oviposition it is our goal to get as accurate a measure of parasitism as possible. Three instances of superparasitism occurred in wheat, with 2 aphids containing 2 larvae each and 1 aphid

containing 3 larvae. Because the rate was so low, superparasitism was included in the counts of complete parasitism. The total parasitism rates are a good estimate, but do not necessarily indicate what would be witnessed in natural populations. To better understand *D. rapae* and their ability to suppress aphid pests in canola in Oklahoma further testing needs to be done under field conditions.

The rates we recorded for *D. rapae* emerging from canola and going into canola are about average when compared to other results (Lopez et al. 1990; Pike et al. 1997; and Fathipour et al. 2006). The complete parasitism rate is a better count of actual parasitism because it is representative of the *D. rapae* larvae that will likely survive to adulthood. These rates are, in some cases, lower than the total but still show that canola is the best emergence and introduced environment for parasitism (Table 1). Because cabbage aphid has been shown to be the preferred host of *D. rapae* it is not surprising that, when available, these have the highest rates (Vaz et al. 2004).

Where this is important is the application of these results to field situations. In wheat, aphid pests are controlled under economic thresholds by the parasitoid wasp, *Lysiphlebus testaceipes* (Cresson) with maximum parasitism rate of 0.2 necessary for complete suppression (Giles et al. 2003). A relatively new crop, canola, offers the opportunity for growers to suppress diseases, treat weeds, and increase wheat yields when used in rotation (Boyles et al. 2009). As canola has begun to be planted in greater acreage, growers are concerned with the aphid pests attacking the crop (Franke et al. 2008) and it is hoped that populations of *D. rapae* already present will move into these fields.

It appears that the initial populations moving from wheat into canola will face a severe reduction in successful parasitism that could hinder their establishment. Because canola is not typically planted for multiple years, the constant re-introduction from wheat could limit the parasitoid's ability to act as an effective biological control agent.

If, instead, *D. rapae* is moving in from wild mustard plants into canola fields, this decrease will not be seen. Baer et al. (2004) found that gene flow occurred readily between native and introduced populations of *D. rapae* and that cabbage aphids are likely the ancestral host and movement into Russian wheat aphid (*Diuraphis noxia* Kurjumov) occurred early on as well. Current populations of *D. rapae* that are found more commonly on Russian wheat aphid are the result of current shifts as genetic diversity between populations using Russian wheat aphid were not different than those using cabbage aphid. Because Russian wheat aphid is not commonly found in Oklahoma (Brewer and Elliott 2003), there is a greater likelihood that the *D. rapae* found in canola are not from the recent populations existing on Russian wheat aphid.

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Table 1. Average Parasitism Rates $\pm$ SE of *D. rapae* for Total, Complete, and Incomplete Parasitism

	Canola/CA to Canola/CA	Canola/CA to Wheat/BCOA	Wheat/BCOA to Canola/CA	Wheat/BCOA to Wheat/BCOA
Total	0.255 ± 0.020	0.005 ± 0.003	0.149 ± 0.019	0.0615 ± 0.012
Complete	0.157 ± 0.015	0.005 ± 0.003	0.103 ± 0.020	0.047 ± 0.009
Incomplete	0.097 ± 0.011	0.00 ± 0.00	0.045 ± 0.017	0.014 ± 0.006

CHAPTER V

SUMMARY

The changes in crops present in the winter landscape in Oklahoma over the past ten years have presented new challenges for pest control. Although a sophisticated system of integrated pest management techniques involving native parasitoids and their aphid hosts is present for wheat, *Triticum aestivum* (Giles et al. 2003), a similar system has not yet been developed for canola (*Brassica napus*).

Although canola has been planted in rotation with wheat since the early 2000's there is still a concern from growers about the pests found in canola, namely cabbage aphid (*Brevicoryne brassica*), turnip aphid *(Lipaphis pseudobrassicae*) and green peach aphid (*Myzus persicae*) (Franke et al. 2008 and Boyles et al. 2009). Because the dominant wheat parasitoid wasp, *Lysiphlebus testaceipes* (Cresson), does not utilize either turnip aphid or cabbage aphid in canola as a host (Elliott et al. 2014) it was important to begin evaluating a different parasitoid for its potential as biological control. The parasitoid *Diaeretiella rapae* (M'Intosh) is known to parasitize all of the aphids present in canola and is thought to use these hosts preferentially to other, non-cruciferous feeding hosts (Read et al. 1970, Pike et al. 1997, Baer et al. 2004, and Vaz et al. 2006).

Diaeretiella rapae is also found in low densities in wheat and is capable of using bird cherry-oat aphid (*Rhopalosiphum padi* L.) as a host. By comparing laboratory populations from both environments we were able to get a more complete picture of what could be happening in the field and the effects different environments can have on a single species.

After separating *D. rapae* from its mummy while it was a teneral adult it became apparent that the chemicals associated with the mummy were crucial to *D. rapae*'s ability to then recognize and find its hosts and their habitats. Un-mated *D. rapae* also showed almost no response to their hosts, instead they responded to both the host plants and feeding volatiles of bird cherry-oat aphid on wheat and cabbage aphid on canola. Our olfactometer tests indicated that *D. rapae* that came from canola (under normal conditions) were more responsive to feeding volatiles than those from wheat.

After comparing rates of parasitism of *D. rapae* we were able to determine that it wasn't just those wasps coming from canola, but those that were searching in canola that had a greater effect on their success (14.8 and 25.5%). Those that came from wheat and were placed into wheat to parasitize bird cherry-oat aphids had a lower rate (6.2%) than either those taken from wheat or from canola and moved into canola. It is the extremely low rate of parasitism in wheat after emerging from canola (0.5%) that could prove to be the most challenging obstacle to using this parasitoid as a biological control agent.

Canola is planted in rotation with wheat and in much smaller acreages-about 0.2 million acres of canola to 6 million acres of wheat annually (NASS 2014). This means that year-to-year the patches of canola will be changing in the landscape and returning to

wheat, a situation that yields almost zero parasitism. In addition to this, *D. rapae* operates

within fields, whereas the main wheat parasitoid, *Lysiphlebus testaceipes*, operates on a

larger scale (Brewer et al. 2008). The likelihood that small populations of *D. rapae* that

have increased in canola will be driven locally extinct when wheat is planted in the same

field the following year increases under these conditions.

Unfortunately the habitat and hosts *D. rapae* uses outside of the canola growing

season are not well known. Conservation of these may prove beneficial to maintaining

populations of *D. rapae*. Further testing of how *D. rapae* reacts to hosts and host plants in

the field is necessary to better understand whether this parasitoid will prove capable of

suppressing, or decreasing, aphid populations.

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