A Survey of Aphid Species and their Associated Natural Enemies in Fraser Valley Hop Fields and an Exploration of Potential Alternative Summer Hosts of the Damson-Hop Aphid, *Phorodon humuli* (Hemiptera: Aphididae)

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ABSTRACT

Six hop fields, *Humulus lupulus* (Rosales: Cannabaceae), in the Fraser Valley, British Columbia, were surveyed throughout 2019 to determine the composition of aphid species and their associated natural enemies. The host range of a known aphid pest of hops, the damson-hop aphid, *Phorodon humuli* (Hemiptera: Aphididae), was also explored in order to further clarify its summer host range and lifecycle. *Phorodon humuli* collected from *Prunus* spp. were reared and transferred to potential alternative hosts including *Cannabis sativa* (Rosales: Cannabaceae) and nettle, *Ultica dioic* (Rosales: Urticaceae).

Phorodon humuli was the dominant aphid species found in all six hop fields in 2019, comprising 99% of all aphids found. *Phorodon humuli* were found in hop fields from May 24, 2019 through to November 3, 2019. Other aphid species were present in some hopyards early in the season including *Aphis fabae* (black bean aphid), *Macrosiphum euphorbiae* (potato aphid) and *Brachycaudus helichrysi* (leaf-curling plum aphid). *Phorodon cannabis* (hemp/cannabis/bhang aphid) was not found in any of the six hop fields.

Ladybird beetles (Coleoptera: Coccinellidae) were the most abundant predator found on hop leaves, making up 76% of all aphidophagous predators found in the six hop fields. *Orius* spp. were the most abundant predator found in hop burrs and cones, contributing to 97% of all aphidophagous predators found in the six hop fields. Predator population growth was low compared to aphid population growth.

In host-transfer experiments, *P. humuli* decreased on cannabis and nettle, while increased on hops, indicating that cannabis and nettle are not a preferred summer hosts of *P. humuli*.

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INTRODUCTION

Background and Setting

The Fraser Valley (Fig. 1) in south-western British Columbia (B.C.) measures close to 150 km/ 93 miles from west to east, and 50 km/30 miles from south to north (Gillespie & Quiring, 2006). The Fraser Valley is characterized by a mild growing climate and quality soils that support a diversity of crops (Fraser Valley Regional District, 2017). The area makes up only a small part of the province, yet it is intensively farmed (Fraser Valley Regional District, 2017).

The hop plant, *Humulus lupulus,* (Rosales: Cannabaceae) is a dioecious, herbaceous perennial. The cones from the female plants are an ingredient used in brewing beer. Cultivation of hops in B.C. first began in Saanich on Vancouver Island in the 1860s, and spread to other areas of the province including Squamish, Vernon, Kelowna and the Fraser Valley (Evans, 2004). Experimental plantings of hops began in the Fraser Valley in 1884 (White, 1948). The experimental planting provided the root stock for the initial hopyards in the Fraser Valley (White, 1948).

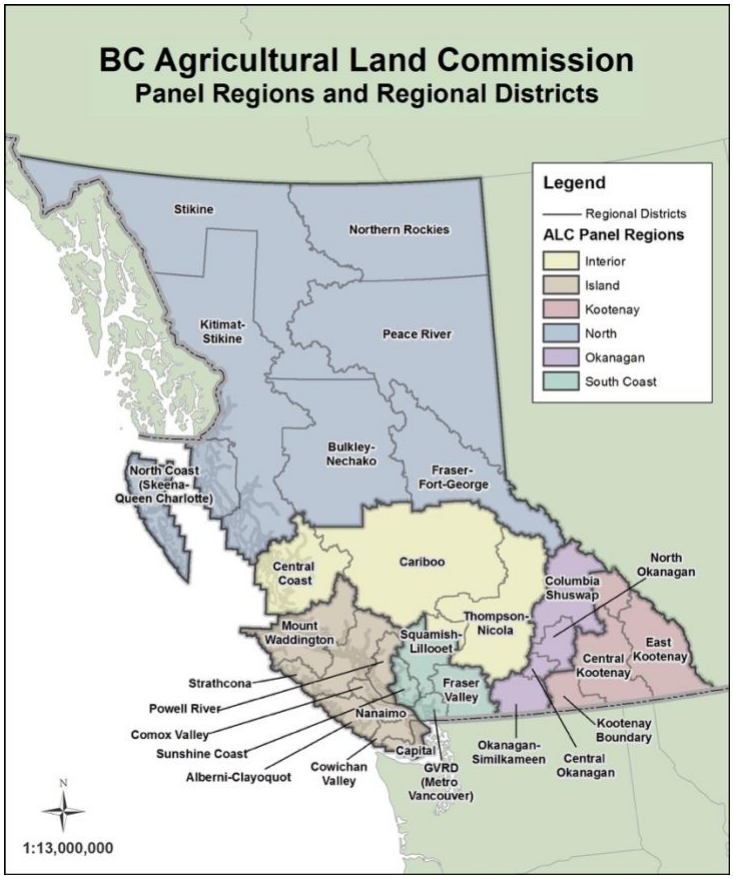


Figure 1: Fraser Valley in British Columbia, Canada (Agricultural Land Commission, 2014)

As other production areas struggled with crop failures due to pests and decreased demands from international export markets, Fraser Valley became the primary growing region of hops in the province (Evans, 2004). During the peak period of hop production in the Fraser Valley in the 1940s (Denman, 2008), there were over 809 ha/2000 acres planted, making it the largest hop-growing region in all of the British Common Wealth (Evans, 2004). Commercial hop production took place in the Fraser Valley from 1893-1997 (Denman, 2008). From 1997, when the last of the large hop operations in the Fraser Valley shut down (Evans, 2004), until approximately 2010, hops were not grown commercially in this region.

Currently, with the onset of micro-breweries, there has been renewed interest in growing hops in the Fraser Valley. There are approximately 80 ha/200 acres of hops currently being grown in this region (Personal communication with Sam Glasgow, Vice Chair, British Columbia Hop Growers Association). Washington state, which neighbours B.C. to the south, currently grows over 16, 187 ha/40,000 acres of hops, making up 73% of the hops produced in the United States of America (U.S.A.) in 2018 (USDA National Agricultural Statistics Service, 2019).

Phorodon humuli (Hemiptera: Aphididae) is commonly known as the 'damson-hop aphid' or the 'hop aphid'. In historic records, *P. humuli* was commonly referred to as the 'hop louse' or 'hop plant louse' (Evans, 2004; Denman, 2008). The damson-hop aphid is often described, along with the two spotted spider mite, as being the major arthropod pest of hops in North America (Campbell, 1978; Dorschner & Baird, 1988; Morrison, 1958; Sirrine, 2009; Turner, Benedict, Darby, Hoagland, Simonson, Sirrine & Murphy, 2011; Weihrauch, 2005; Woods, Dreves, James, Lee, Walsh & Gent, 2014; Wright, Pike, Allison & Cone, 1995). *Phorodon humuli* has a host-alternating lifecycle (Calderwood, Lewins, & Darby, 2015; Cranham, 1982; Lösel, Lindemann, Scherkenbeck, Campbell, Hardie, Pickett & Wadhams, 1996; Pérez, Seco, Valenciano, 2007; Wright, Cone & James, 2005; Wright & James, 2001).Various *Prunus* spp. (Rosales: Rosaceae) are used as the winter or primary host and hops, *Humulus lupulis*, (Rosales: Cannabaceae) are listed as the summer or secondary host (Campbell & Cone, 1994; Cranham, 1982; Lorenzana, Hermoso de Mendoza, Seco & Casquero, 2010; Lösel et al., 1996; Morrison, 1958; Pérez et al., 2007; Wright et al., 1995, 2005; Wright & James, 2001).

Phorodon humuli can be economically problematic for the hop industry for several reasons. Infestations of *P. humuli* can reduce the number of cones, weaken plants, and slow bine growth (Lorenzana et al., 2010). Left uncontrolled, *P. humuli* can defoliate the hop plant (Campbell & Cone, 1994). *Phorodon humuli* can also vector *Hop mosaic virus, American Hop Latent Virus* and *Hop Latent Virus* (Eastwell & Barbara, 2015). Of particular concern is the aphid's excretion, or honeydew, as it serves as an ideal medium for the development and growth of sooty mold (*Cladosporium* spp.) (Pike & Starý, 1995). The presence of sooty mold in cones can result in a downgraded product or a complete crop rejection (Campbell & Cone, 1994; Dorschner & Baird, 1988).

In the Fraser Valley, *P. humuli* was present in the late 1930s (Fig. 2). The fall of the hop industry on Vancouver Island has been attributed to hop louse (i.e. the damson-hop aphid) (Denman, 2008), which also serves as evidence of this pest in the province. While there are historic records of *P. humuli* in British Columbia (Fig. 2; Denman, 2008), no recent work has been done to

confirm the current presence of *P. humuli*, or to identify any other aphid species present in hop fields in the Fraser Valley.

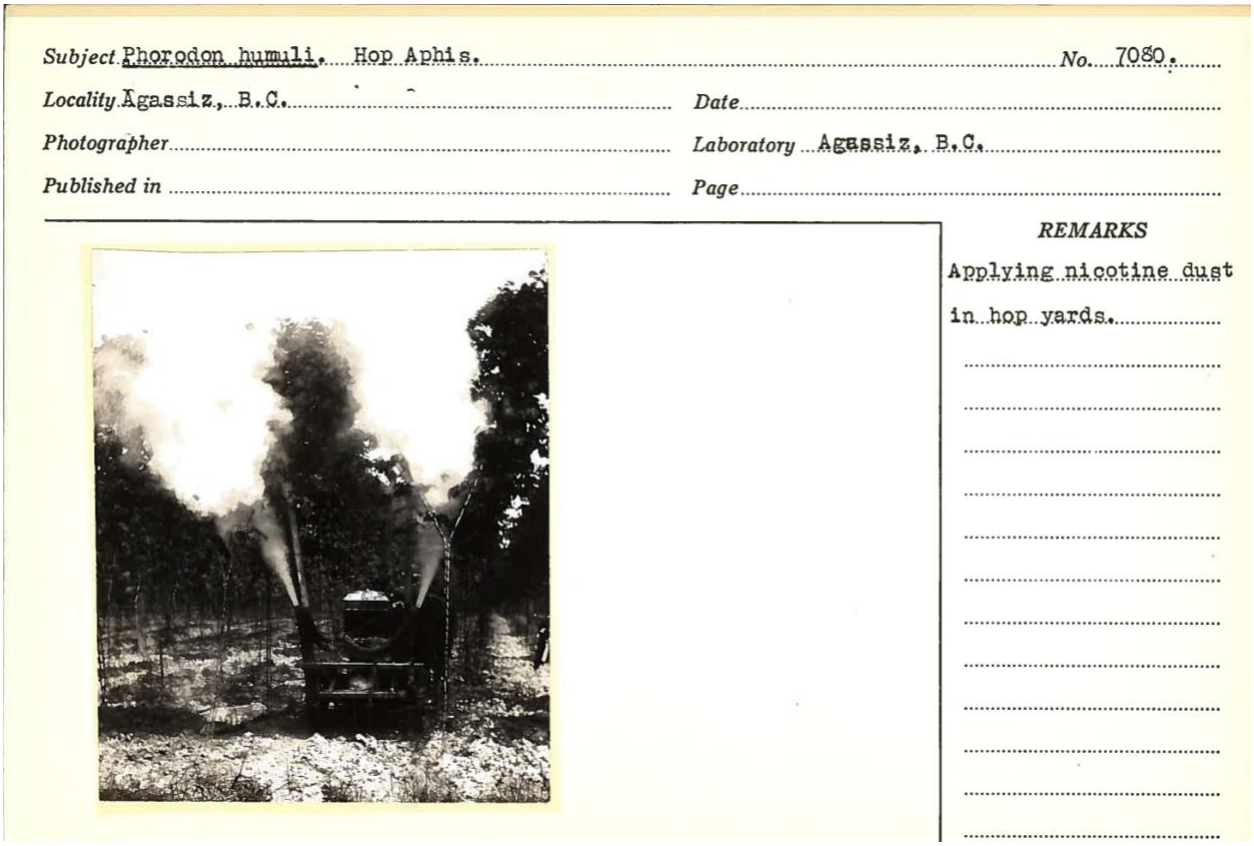


Figure 2: Historic record of *Porodon humuli* in Fraser Valley. Photo courtesy of Paul Abram, Agriculture and Agri-Foods Canada.

Unlike the historic years, the new era of hop growing faces new potential pest pressure from a closely related crop. *Cannabis sativa* (Rosales: Cannabaceae) was legalized in Canada in October of 2018, and plantings of outdoor cannabis and hemp have since ensued. Residents in B.C. are allowed to grow up to four cannabis plants per household, and these plants can be grown either outdoors or indoors (Province of British Columbia., n.d). Both hops and cannabis are in the same botanical family, Cannabaceae. Hammon, Rizza, & Dean (n.d.) have highlighted potential pest sharing between these crops. The authors state that "Hops is the plant species that faces the greatest potential crossover threat from any of the Cannabis specialist insects. Conversely, Cannabis is the plant species that faces the greatest crossover threat from any of the hop specialist insects" (Hammon et al., n.d., p. 1). Both large scale commercial production of cannabis, as well as many pockets of small-scale homegrown cannabis, have the potential to change the pest dynamics in contemporary Fraser Valley hop production. *Phorodon cannabis* (Hemiptera: Aphididae), which is commonly referred to as the bhang aphid or the cannabis aphid, is a pest of particular interest.

Statement and Significance of the Problems

1) There is currently a lack of understanding of the species composition of aphids in hop fields in the Fraser Valley. Since its revival as a crop in this area, region-specific research on pest management has been minimal. Hop growers in this region are unfamiliar with the species present in their fields. There is no hop production guide for the Fraser Valley which describes common pests of this crop in this particular region. While there are historic records of *P. humuli* in this region (Fig. 2), there is a need to confirm its presence and status in current hop production in the Fraser Valley.

2) *Phorodon humuli* is listed as the only aphid pest in hop growing guides from northeastern U.SA. (Calderwood et al., 2015), northwestern U.S.A including Washington, Oregon and Idaho (Dreves & Walsh, 2015), and Michigan (Lizotte, Hodgson & Filotas, 2017). Considering the diverse agricultural production in the Fraser Valley, and the diverse lifecycles of aphid species, there is a need to document if there are other important aphid species present in hop fields in this region. While large hop-growing regions to the south of the Fraser Valley, in Washington state, can provide some insight into the aphid species and predator composition to be expected in the Fraser Valley fields, region-specific pest management knowledge (that reflects the influence of both climate and surrounding crops, such as cannabis), is required for successful pest management.

3) There are contradicting statements in the literature regarding the general life cycle and summer host range of *P. humuli* (McPartland, Clarke & Watson, 2000; Smith, 1951; Campbell, 1985) especially in light of cannabis and hemp production, crops that share botanical similarities to hops. Confirming any potential secondary host of *P. humuli*, including cannabis, is important for understanding its potential spread to and from other commodities grown regionally. Alternative summer hosts besides hops could have an impact on population levels in hop fields, timing of when they enter into hop fields, and control measures taken.

4) Current management of aphids in hops in the Fraser Valley is primarily achieved through chemical control. Four active ingredients are registered for control of aphids in this crop (Pest Management Regulatory Agency Online Label Search, n.d.). An integrated pest management framework is needed to ensure continued control of the pest, and product stewardship in the future. Understanding the natural enemies of aphids in hop fields is the first step in understanding how to take an integrated approach to managing aphids in hop fields. Currently no regional information is available.

Purpose of the Project

The purpose of the project is to contribute to the body of knowledge surrounding pest management of hops in the Fraser Valley by surveying the aphid species present in hop fields, their associated natural enemies, and confirm the host range of a known pest of hops, *P. humuli.*

Project Objectives

Objective 1: To determine aphid species composition in hop fields in the Fraser Valley, British Columbia.

Objective 2: To determine the associated aphidophagous predators and parasitoids in hop fields in the Fraser Valley, British Columbia.

Objective 3: To study the effect of summer plant host on the ability of *P. humuli* to survive and produce new aphids.

LITERATURE REVIEW

Phorodon humuli Phenology and Lifecycle

Phorodon humuli migrates between *Prunus* spp. in the winter and hops, *Humulus lupulis*, in the summer, and overwinters as eggs on *Prunus* spp. (Calderwood et al., 2015; Morrison, 1958). Eggs are laid on *Prunus* spp. in the fall, and hatch takes place in early spring, during or before bud burst of *Prunus spp.* (Cranham, 1982). Morrison (1958) writes that egg hatch can occur as early as February if the weather is warm, but can be prolonged until late May. In south-central Washington, eggs hatch between February and March (Wright et al., 2005).

Apterous (wingless) females hatch from the eggs laid on *Prunus* spp. (Cranham, 1982). Several more generations of apterous females are then produced viviparously (by live birth) on *Prunus spp.* (Calderwood et al., 2015; Cranham, 1982; Wright et al., 2005). These generations feed on the buds and leaves of *Prunus* spp. (Cranham, 1982). It has been suggested that female alates (winged) are produced in response to decreasing food quality of *Prunus* spp. (Cranham, 1982; Pérez et al., 2007). Cranham (1982) suggests that the development of alates is triggered by the termination of *Prunus* spp. shoot growth in mid-May to June. Accumulated degree days were calculated for the first alatoid aphids (showing features of alates) in Washington and were found at 397 degree-days in 1989 (May 16); 378 degree-days in 1990 (May 10); and 283 degree days in 1991 (May 7) (Wright et al., 1995).

The alates of *P. humuli* that are produced on *Prunus* spp. can begin their migration to hops when daily temperatures reach 13 °C (Calderwood et al., 2015; Cranham, 1982). In England, Cranham (1982) suggested migration to hops begins at the end of May, and continues until mid-July, occasionally into August. In a three year study in England, spring migration of *P.humuli* varied yearly, from mid-May until early June (Aveling, 1981).The timelines for spring migration to hops are similar in Washington: Campbell & Cone (1994), stated that migration to hops occurs in late May, however, it has been noted to start as early as the beginning of May (Wright et al., 1995). In Oregon, Morrison (1958) dated the migration to hops in late May. In order to understand the end of migration to hops, accumulated degree days were recorded in Washington to mark the date the last alatoid aphids were found on *Prunus* spp. in the spring/summer (Wright et al., 1995). The accumulated degree days varied over the three years, but the mean accumulated degree days for the last alatoid found on *Prunus* spp. was 1,222 (Wright et al., 1995). Wright et

al. (1995), but concluded that many more years of data were necessary to accurately test the accumulated degree day.

Once the female alates (winged) arrive to the hops, they begin to produce parthenogenetic viviparous apterous females, and this form of reproduction carries on throughout the summer (Campbell, 1977, 1985; Cranham, 1982; Pérez et al., 2007; Wright et al., 2005). It is suspected that there are around 10 overlapping generations per hop season in the Pacific Northwest, and each female can produce 30-50 nymphs in her two-four week lifecycle (Dreves & Walsh, 2015). Winged females are not produced during the summer on hops (Campbell, 1985; Cranham, 1982).

In the fall, both winged males and female gynoparae (parthenogenetic viviparous aphids that are responsible for producing the sexual females) develop on hops to fly back to their winter host, *Prunus* spp. (Pérez et al., 2007). The flight of both males and females back to *Prunus* spp. from hops has been connected to shortening day length hours (Cranham, 1982). In Washington, Wright et al. (1995) found the timing of the fall migration to Prunus can be triggered by both decreasing day length and temperature. Migration to Prunus started at 13.5 hours of day length (Wright et al., 1995). According to Wright et al. (1995), the fall migration can start earlier (when days are longer than 13.5 hours), if there are extended periods of cooler temperatures (less accumulated degree-days). In Washington, the end of the fall migration of *P. humuli* back to *Prunus* spp. is marked by frost-killed foliage on hops (Wright et al., 1995).

Once back on *Prunus* spp., the winged females produce wingless oviparae (female aphids that lay eggs instead of giving birth to live young) (Pérez et al., 2007; Wright et al., 1995). When the winged males arrive to *Prunus* from hops, they mate with the egg-laying wingless females (Wright et al., 1995). Wright et al. (1995) suggest that the high ratio of females to males indicate that males mate with more than one female. After mating, eggs are laid in the bud axils (Cranham, 1982; Taylor et al., 1979) to remain for the winter and reproductive male and females die.

Flight

The distances travelled by *P. humuli* are thought to differ between spring and fall migration (Loxdale, Brookes, Wynne & Clarke, 1998). There is difficulty in determining flight distances of aphids due to their size and ability to use air currents and wind to aid in dispersal (Taylor, Woiwood & Taylor, 1979). Loxdale et al. (1998) suggests that the migration in the fall is shorter, and probably no more than 20 km. For fall migration, Taylor et al. (1979) suggest the median distance travelled by *P.humuli* to be 15-20 km if aphids reached a flight height of 12.2 m. For spring migration, Campbell (1977) estimated a flight distance for *P. humuli* of 16-32 km. Taimr & Kriz (1978) found *P.humuli* 22 km from the initial Prunus source.

Winter Hosts of *Phorodon humuli*

The genus, *Prunus*, includes plums, cherries (*Prunus avium* and *Prunus cerasus*), peaches (*Prunus persica*), nectarines (*Prunus persica* var. nucipersica), apricots (*Prunus armeniaca*),

and almonds (*Prunus dulcis*). *Phorodon humuli* can use any of those as winter hosts, but it's preferred host is plums, both ornamental and fruit bearing (Wright et al., 2005, 1995). Much of the information on winter hosts of *P. humuli* comes from England and Washington.

Both ornamental and fruit plums are used as hosts by *P. humuli* (Wright et al., 2005, 1995), including *Prunus cerasifera* Ehrhart, *Prunus divaricate, Prunus domestica, Prunus insititia, Prunus mahaleb* and *Prunus spinosa* (Write et al, 2005).

In England, the sloe or blackthorn, *Prunus spinosa*, serves as the primary host of *P. humuli*. Other, less common hosts, are the damson plum, *Prunus insititia*; the common plum, *P. domestica*; and the cherry plum or myrobalan plum, *P. cerasifera* Ehrh (Cranham, 1982; Taylor et al., 1979).

In Central Washington, the cherry plum, or myrobalan plum, *Prunus cerasifera* Ehrhart cv. Thundercloud has been noted for its importance as an overwintering site for *P. humuli* (Wright & James, 2001). In a multi-year study in Washington, Wright et al. (2005) sampled multiple *Prunus* spp. from small orchards, residential yards, commercial areas and parks, and found that purple-leafed ornamental flowering plums were the primary source of *P.humuli* in the spring. The authors attribute this importance to the declining plum and prune industry in the area, and because trees in commercial fruit orchards are often sprayed (Wright et al., 2005).

The genus, *Prunus*, is not only important for a source of spring migrants to hop field, but the penology of the plant has been used in attempts of developing a biofix for accumulating degree-days for *P. humuli.* Based on historical records in England, Worner, Tatchell, & Woiwod (1995) were able to confirm that egg hatch of *P.humuli,* is connected with bud burst of different *Prunus* (Worner et al., 1995).

Summer Hosts of *Phorodon humuli:* Discrepancies in the Literature

Most often, hops are listed as the only secondary (or summer) host of *P. humuli* (Campbell, 1985; Campbell & Ridout, 2001). Wright et al. (1995) suggest that hops are the only "important secondary host" of *P. humuli* (p.10). However, there are two references to nettle as a host of *P. humuli:* Smith (1951) lists nettle, *Ultica dioic,* as another host of *P. humuli;* and Taylor et. al (1979), states "If hop aphids feed on nettles, as Patch (1938) suggested, they must do so very rarely, for nettles are ubiquitous and autumn hop aphids are not" (p. 962).

Phorodon humuli has also been listed as an aphid pest of cannabis. McPartland et al. (2000) list six aphids present on *Cannabis* including *Myzus persicae* (green peach aphid), *Aphis fabae* (black bean aphid), *P. cannabis* (hemp/cannabis/bhang aphid), *P. humuli* and *Aphis gossypii* (melon or cotton aphid). Mcphartland et al. (2000) also cites three authors that have observed *P. humuli* on cannabis: Blunch (1920), Flachs (1936) and Eppler (1986). Unfortunately, none of these studies are in English, so the statements could not be confirmed from the source material. Blackman and Eastop (1984) do not list *P. humuli* under aphids found in hemp. Hammon et al. (n.d.) have flagged *P. humuli* as an established pest specialist in Colorado that has potential to be a pest of outdoor-grown cannabis. In a 1989 survey of insects associated with outdoor cannabis

in Mississippi, the only aphid noted was *Hysteroneura setariae* (Thomas), commonly known as the rusty plum aphid (Lago & Stanford, 1989).

Other Aphids on Hops

The available field guides list *P. humuli* as the sole aphid species for hops (Dreves & Walsh, 2015; Lizotte et al., 2017). In Oregon hopyards, Woods, et al. (2014) did not note any additional aphid species besides *P. humuli*. Campbell and Cone (1994) state that it is uncommon for other aphid species to feed on hops.

While Blackman and Eastop (1984) do not list *P. humuli* under aphids found in hemp, they do list *P. cannabis* under aphids found on hops. This observation has also been noted by Cranshaw et al. (2018), but the authors take the stance that *P. cannabis* is monophagous on Cannabis. In addition to *P. cannabis*, Blackman and Eastop (1984) list *Aphis fabae, Aphis (Cerosipha) humuli, Aphis gossypii, Phorodon humuli foliae, Phorodon humuli humuli, Phorodon humuli japonensis, Rhopalosiphoninus staphyleae, Myzus persicae, Aulacorthum solani, Macrosiphum euphorbiae* and *Macrosiphum hamiltoni* as other aphid species found on hops.

Hop Cultivar Susceptibility to *Phorodon humuli*

The susceptibility of certain commercial hop cultivars to *P. humuli* has been determined for many European cultivars. In a German hop cultivar preference study, 'Hallertauer Magnum,' was found to be more susceptible to *P. humuli* compared to 'Spalter Select' in field studies where both spring migration numbers and population development were examined (Weihrauch & Moreth, 2005). In this same study, alates of *P. humuli* were put into plants in a greenhouse, and aphids exhibited more probing and searching behavior on 'Spalter Select' compared to 'Hallertauer Magnum' (Weihrauch & Moreth, 2005). After 28 days in the greenhouse, more aphids developed on 'Hallertauer Magnum' compared to 'Spalter Select' (Weihrauch & Moreth, 2005).

In England, Campbell (1977) looked at the influence of both cultivar and wind shelter as factors relating to the distribution of *P. humuli* in hops. More migrant hop aphids arriving into hop fields were found on 'Northern Brewer' compared to 'Tolhurst' (Campbell, 1977). The three cultivars studied were ranked from least to most susceptible to incoming migrant aphids: 'Tolhurst' was the least susceptible, 'Fuggle' was moderately susceptibility and 'Northern Brewer' was the most susceptible (Campbell, 1977). In a later study, the same three cultivars ('Tolhurst,' 'Fuggle' and 'Northern Brewer') were examined in relation to the reproduction, development and survivability of *P. humuli* (Campbell, 1983). 'Northern Brewer' was found again to be the most susceptible cultivar (Campbell, 1983). Campbell (2018) used 'First Gold' and 'Herald' as aphid-susceptible cultivars in a companion planting study.

A breeding line 23/90/08 was developed from a male plant in Japan that had aphid-resistant genes, and has been tested against 'First Gold' and 'Herald,' which are other aphid susceptible cultivars (Barber, Campbell, Crane, Darby, & Lilley, 2003). Its resisitance to aphids was found to have similar effects to one pesticide application (Barber et al., 2003). This breeding line was

used in the development of a dwarf cultivar, 'Boadicea,' and has since been used as an aphid-resistant cultivar by Campbell (2018).

In the U.S.A, Dorschner & Baird (1988) screened for *P. humuli* resistance in the hop genotypes from the United States Department of Agriculture World Collection of Hop Geneotypes. Hop genotypes showing *P. humuli* resistance were compared to the resistance in commercially grown hop cultivars in lab experiments where aphids were caged on plants (Dorschner & Baird, 1988). Out of the commercial cultivars, 'Cascade' was most susceptible to *P. humuli* and 'Perle' was the least susceptible (Dorschner & Baird, 1988). The rate of growth of *P. humuli* on 'Perle' was significantly lower compared to the other commercial cultivars tested, except for 'Chinook,' indicating 'Chinook' has some resistant traits (Dorschner & Baird, 1988).

There has also been breeding work done to create hop cultivars that are resistant to *P. humuli.* Interest in breeding hop cultivars resistant to *P.humuli* has not always been a priority. This, in part, could be attributed to the availability, success and efficacy of chemical pesticides (Campbell, 1983; Dorschner & Baird, 1988). An emphasis on breeding programs for resistant cultivars to fungal diseases also took priority over the development of varieties resistant to aphids due to the availability of effective aphicides (Campbell, 1983).

Thresholds

The bitterness of hops is an important property in brewing, and is measured by the quantity of alpha acids (Lorenzana et al., 2010). In attempts to understand an economic threshold for *P. humuli,* aphid levels were examined in connection with alpha acid levels (Lorenzana et al., 2010; Weihrauch, Baumgartner, Felsl, Kammhuber, & Lutz, 2012). Lorenzana et al. (2010), showed that aphid density in cones did not impact alpha-acid levels. In Weihrauch et al. (2012), aphid levels impacted alpha-acids and yield differently each year. In situations where aphid levels were high within the cones, but did not negatively impact the quality standards by which the authors were assessing the hops, the hops were downgraded to the point where they would be unsellable by visual standards (Weihrauch et al., 2012). So, in addition to brewing characteristics, hop quality is also impacted by aesthetics. Turner et al. (2011) summarize the complexities: "Due to the direct correlation between quality and price of hops, a crop can be drastically affected by pests and diseases that alter not just the brewing quality but also the aesthetics of the crop as well. Any loss of quality can cause a crop to lose value or be damaged to the point at which it is completely unsalable" (p.1647). It is difficult to assess the point at which *P. humuli* causes economic damage to the crop due to the various parameters in which hop quality is assessed. While there are no economic thresholds for hop aphids (Woods et al., 2014), working thresholds are commonly used that take account of the complexities of the presence of hop aphid in relation to quality, aesthetics and yield.

There is an emphasis on controlling *P. humuli* prior to cone formation in order to prevent aphids from entering into the cones (Aveling, 1981; Dreves & Walsh, 2015). Once in the cones, aphid feeding leads to the development of sooty mold (Campbell & Cone, 1994). Additionally, once in the cones, aphids are protected from both contact pesticides (Aveling, 1981) and many natural enemies (Campbell & Cone, 1994). As a result, the working thresholds that are used focus on the

number of aphids per leaf. The provisional or working thresholds that have been used in place of economic thresholds are similar across North America. In Michigan, pesticide application is recommended at 8-10 aphids per leaf prior to cone development (Sirrine, 2009). In Oregon, a working threshold of 5-10 aphids per leaf prior to cone development is used (Dreves & Walsh, 2015; Woods et al., 2014). In Ontario, management for hop aphids with insecticides is simply recommended "before pest populations become too large" (Filotas, 2012, p.34). In northeast U.S.A., an economic threshold does not exist (Calderwood et al., 2015).

Predators

There has been considerable effort in identifying and documenting the important groups of predators that contribute to biological control of *P. humuli* in hopyards (Aveling, 1981; Barber et al., 2003; Calderwood et al., 2015; Campbell, 1978; Campbell & Cone, 1994; Copland, 1979; Lorenzana et al., 2010; Woods et al., 2014). Of those groups, Coccinellidae (ladybird beetles), predatory Hemiptera, Neuroptera (lacewings) and predatory Diptera will be the focus of the review.

Coccinellidae (Ladybird Beetles)

The coccinellids, the ladybird beetles, have been documented for their control of aphids and other soft bodied arthropods in many crop settings. Campbell and Cone (1994) suggest that that Coccinellids were the most important predator in hopyards because of their abundance and ability to eat many aphids. The most commonly occurring species of coccinellids in Campbell and Cone (1994) were *Coccinelle transversoguttuta,* followed by *Hippodamia convergens*. *Adalia bipunctata* was also found (Campbell & Cone, 1994). In a nine-year study in newly planted Oregon hop fields, coccinellids were present every year and included the following species: *Harmonia axyridis*, *Cycloneda polita, Coccinella septempunctata, Coccinella transversoguttata, Adalia bipuntata* and *Psyllobora* spp. (Woods et al., 2014).

In hopyards in England, ladybird beetles have not been the most abundant predator, however, they are often the first predators to arrive to the field in the spring (Barber et al., 2003; Campbell, 1978). Aveling (1981) found four species of ladybird beetles including *Adalia bipunctata, Coccinella undecimpunctata, C. septempunctata, and Propylea quatuordecimpunctata*, however, the author noted that in all years of the three year study, their numbers were low when compared to anthocorids (Aveling, 1981).

Predatory Hemiptera

While the composition of species differs between fields in Europe and in North America, the family Anthocoridae, often called minute pirate bugs or flower bugs, plays an important role in the predation of *P. humuli* (Aveling, 1981; Campbell, 1978; Campbell & Cone, 1994; Woods et al., 2014).

In England, Campbell (1978) found that earwigs and anthocorids were the only abundant natural enemies present where *P. humuli* levels were reduced. Seasonally, anthrocorids were the most abundant predator from July onwards (Campbell, 1978). *Anthocoris nemorum* and *A. nemoralis*

were two species that were regularly found, whereas *A. confuses* and *Orius majusculus* were found less often (Campbell, 1978). In a three-year field study in England, anthocorids of the genus *Anthocoris* were the most common predators found, and contributed the most to the predation of *P. humuli* (Aveling, 1981). Of all the *Anthocoris* spp., *Anthocoris nemoralis* was found most frequently each year (Aveling, 1981). The findings of Aveling (1981) show that the decline of *P. humuli* corresponded with the peak population of the fourth and fifth nymphal instars of anthocorids. Additionally, anthocorids were the only predators found in cones (Aveling, 1981).

In another three-year study in Spain, Lorenzana et al. (2010) found that *Orius* spp. were the most commonly found natural enemies of *P. humuli* in cones. *Orius* spp. are generalist predators which feed on eggs, larvae, and adults of various other species of agricultural pests (Herring, 1966). *Orius* spp. also feed on pollen and mites, and are quite mobile (Lorenzana et al., 2010). Over the course of nine years, anthocorids including *Orius* spp. were present on leaves in Oregon hop fields every year (Woods et al., 2014). In Washington, *Orius tristicolour* was the most common anthocorid found in hopyards, but was low in abundance compared to chrysopids and coccinellids (Campbell & Cone, 1994). In Washington, *Nabis* spp. (Hemiptera: Nabidae) were the second most abundant Hemiptera found after *Orius* spp.(Campbell & Cone, 1994). *Nabis* spp. were not, however, found on leaves, but rather were found via beat sampling (Campbell & Cone, 1994). *Nabis* spp. were also found in Oregon hop yards, however, were not found during all nine years of the study (Woods et al., 2014). Big eyed bugs, *Geocoris* spp. (Hemiptera: Geocoridae) were not listed in the predatory Hemiptera found in Oregon hopyards (Woods et al., 2014), but were found in Washington fields (Campbell & Cone, 1994) although their abundance was not noted.

Neuroptera: Hemerobiidae & Chrysopidae (Lacewings)

In Oregon, lacewing larvae (Neuroptera: Chrysopidae, Hemerobiidae) were listed as one of three key beneficial arthropods occurring on hops (Walsh, 2015*). Chrysopa* spp., *Chrysoperla* spp., and *Hemerobius* spp. are said to commonly occur in hop fields (James & Dreves, 2015), however Woods et al. (2014) found low levels of both green and brown lacewings. In Washington, *Chrysoperla carnea* and *C. rufilabris* were commonly found (Campbell & Cone, 1994). On dwarf hop varieties in England, Chrysopidae were not seen (Barber et al., 2003) while in hopyards of standard height in other English hop fields, *Chrysoperla carnea* was found in low numbers every year over the course of a three year study (Aveling, 1981).

Predatory Diptera

Predatory Diptera, such as Syrphidae (syrphid flies) and Cecidomyiidae (gall midges, in particular, *Aphidoletes* spp.) are often associated with aphid predation (Patterson & Ramirez, 2016; Van der Ent, Knapp, Klapwijk, Moerman, van Schelt, de Weert, 2017). Syrphid flies are common predators found in annual cropping systems in the south-coast of British Columbia (Prasad, Kabaluk, Meberg, Bevon & Henderson, 2009). *Aphidoletes* spp. can be purchased from many biological control companies for control of aphids in greenhouses. Both aphidophagous syrphid flies and gall midges, however, appear to only play a small role in predation of *P. humuli*

in hop fields in Europe (Aveling, 1981; Barber et al., 2003; Campbell, 1978) and in North America (Campbell & Cone, 1994; Woods et al., 2014).

Other Natural Enemies

In addition to predators, parasitoids of *P. humuli* have also been documented on both *Prunus* spp. (Wright & James, 2001) and hops (Pike, Starý, Miller, Graf, Allison, Boydston & Miller, 2000; Pike & Starý, 1995). Aphid parasitoids often rely on specific aphid species for development and survival (Patterson & Ramirez, 2016; Van der Ent et al., 2017).

Aphid-Parasitic Hymenoptera

Worldwide, nine species are listed as parasitoids of *P. humuli* including: *Aphidius ervi*; *Aphidius matricariae*; *Binodoxys conei*; *Diaeretiella rapae*; *Ephedrus persicae*; *Ephedrus plagiator*; *Lysiphlebus testaceipes*; *Praon volucre*; and *Trioxys humuli* (Pike & Starý, 1995).

In hop propagation greenhouses in England, *P. humuli* is parasitized by *Aphidius* spp. (Copland, 1979). In a companion planting study in England hopyards, *Aphidius matricariae* was the primary aphid parasitoid species found (Campbell, 2018). Other primary parasitoids included *Aphidius picipes*, *Ephedrus plagiator*, *E. persicae*, *Trioxy humuli*, *Lysiphlebus fabarum* and *Praon volucre* (Campbell, 2018).

In the northwestern USA, eight primary parasitoids of *P. humuli* (Table 1) have been recorded (Pike et al., 2000). *Binodoxys conei* is described as a monophagous parasitoid of *P. humuli* in Washington (Pike & Starý, 1995).

Table 1: Summary of primary parasitoids of *Phorodon humuli* categorized by host. Information consolidated from Pike et al. (2000).

Parasitoid name	**Parasitizes *Phorodon humuli* while on *Prunus* spp.**	**Parasitizes *Phorodon humuli* while on hops**
Aphidius ervi	X	
Aphidius matricariae		X
Binodoxys conei		X
Diaeretiella rapae		X
Lysiphlebus testaceipes	X	X
Monoctonus campbellianus	X	
Praon occidentale		X
Praon unicum	X	

In Washington hopyard surveys, aphid parasitoids have been rare (Campbell & Cone, 1994; Pike & Starý, 1995). Similarly, in hopyards in England, Copland (1979) found that populations of aphid-parasitic Hymenoptera did not increase with *P. humuli* populations nor contribute to the control of this pest. Campbell (1978) reported a rate of less than 1% aphid parasitism in his study of natural enemies of *P. humuli*.

While aphid parasitoids in hopyards seem to be rare (Campbell & Cone, 1994; Pike & Starý, 1995), a two-year study conducted in Washington found that hop aphids are frequently parasitized on their overwintering host, *Prunus* spp. (Wright & James, 2001). *Lysiphlebus testaceipes* was the most common primary parasitoid in both years of the study and *Praon unicum* was the second most abundant primary parasitoid (Wright & James, 2001). Hyperparasitoids were very common and outnumbered primary parasitoids in both years of the study (Wright & James, 2001).

Entomopathogenic Fungus

In addition to predators and parasitoids, fungal pathogens can also play a role in the control of aphids. Infection of *P. humuli* by fungal pathogens has been observed in the field, however, the pathogens have not always been identified (Barber et al., 2003). In the field setting, Barber et al. (2003) noted aphids infected by fungi, and observed the highest occurrence of the fungus was found in areas where aphid levels were highest (Barber et al., 2003). There was however, no mention of the specific fungus responsible for this (Barber et al., 2003). In another field study, infection of *P. humuli* by *Entomophthora* spp. was observed in high aphid densities (Campbell, 1978). Dorschner, Feng & Baird (1991) had some success in infecting *P. humuli* with *Beauveria bassiana* in potted plants and in the laboratory, but not in the field where crops were grown in soil.

PROJECT METHODOLOGY

Targeted Population

The targeted audience for this research is hop and hemp growers and their pest advisors.

Participating Audience

Commercial hop growers of the Fraser Valley, and Garside Fruit Farm, where Transfer Experiments 1 & 2 were performed.

1) Pre-harvest Field Survey Study for Aphids and Natural Enemies

Selection and Description of Sites

Six commercial, conventional hop fields throughout the Fraser Valley, British Columbia, were monitored every second week from May 10 until September 13, 2019 (Fig. 3). Fields were selected based on grower willingness and interest to participate. Of the six fields, three were the cultivar, ‘Cascade,’ and three were the cultivar, ‘Triple Pearle’ (Table 2).

Figure 3: Map of six hop sites surveyed in the Fraser Valley, British Columbia (Canada) in 2019

Table 2: Summary of site characteristics of six Fraser Valley hop fields surveyed in 2019 for aphids and natural enemies

						Description of adjacent land surrounding farm									
Farm/site	Total farm acreage	Acres in 2019 study	Year planted	Hop cultivar used in study	Interrow planting description	Hay	Berries	Corn	Poultry	Mixed flower	Mixed veg	Hemp	Forest	Riparian	Golf course/turf
1	9	4.5	2015	Cascade	Perennial grasses	X	X			X			X	X	
2	25	4.2	2017	Triple Pearle	Perennial grasses		X	X						X	
3	20	2	2016	Triple Pearle	Perennial grasses			X	X	X					X
4	6	1	2016	Cascade	Bare		X					X			X
5	12	3.34	2017	Triple Pearle	Bare	X		X			X				
6	6	2	2014	Cascade	Bare		X		X						X

In-Field Sampling for Aphids and Natural Enemies

In all six fields, in-field sampling was conducted from May 10, 2019 until harvest. Harvest dates differed between fields, occurring from late August to mid-September. The last pre-harvest sampling date was on September 13, 2019. Shoots, leaves, burrs and cones were examined for aphids and natural enemies throughout the season at two-week intervals. Sampling changed to reflect plant growth (Table 3). Re-entry times of pesticides were observed, resulting in some missed sampling weeks.

Table 3: Summary of hop plant parts sampled for aphids and natural enemies throughout different times the 2019 season

Part of hop plant sampled	**Sampling Date**									
	May 10	May 24	June 6	June 21	July 5	July 19	Aug. 2	Aug. 16	Aug. 30	Sept. 13
Shoots (in field)	X	X								
Leaves (removed from field)		X	X	X	X	X	X	X	X	X
Burrs/Cones (removed from field)						X	X	X	X	X

Each week, two rows per field site were selected randomly. Five plants were selected per row; one at each row end, and three evenly spaced. Per field, 10 plants were selected, four of which were plants at row ends. For the first two weeks (May 10 and May 24, 2019), one edge row and one middle row were selected.

From each plant, five leaves and five cones were taken. Leaves and cones were taken from various heights on the plant. The hop bine canopy was divided in five vertical sections: ground, lower, mid, high and top. One leaf and one cone were taken from each section for a total of five leaves and five cones per plant. This resulted in 50 leaves per field and 50 cones per field per week. As burrs appeared and turned into cones, samples were taken as available. This did not equate to five burrs per sample until week 7 (August 2, 2019).

Two pruners were used throughout the season to ensure leaves and cones were sampled from all height levels. Corona LR 3460 Long Reach Cut 'n' Hold Bypass Pruner (117 cm/46 inches) was used for lower-mid canopy sampling throughout the 10 weeks. Fiskars Chain Drive Extendable Pole Saw & Pruner (2-4.8 m/7–16 ft) was used starting June 7 (week 3) and in subsequent weeks to ensure samples were taken up to 6.4 m./21 ft (Fig. 4).

Pruners were cleaned between each field with rubbing alcohol (70%) and disposable paper towel. At the end of each sampling day, pruners were placed into a bucket of hydrogen peroxide overnight.

Figure 4: Fiskars pruners used for sampling leaves and cones for aphids and natural enemies

During the first two weeks (Table 3), shoots were not removed as this would interfere with apical growth and normal production practices. If aphids were found on shoots, they were recorded in the field and collected in 70% ethanol to later confirm identification.

Leaves and cones were collected and placed into plastic bags and identified out of the field. If they could not be counted and identified on the day of the field visit, they were stored in a refrigerator at approximately 4°C/39°F for up to one week. Aphids and predators on leaves and cones were examined, counted and recorded onto a data sheet. Data was then transcribed electronically.

Identification of Aphids

Aphids were identified to the species level. Aphids were examined for identifying features under a digital microscope, Dino-Lite Edge 1.3MP WF4115ZT. Two keys were used to identify aphids. The key for aphids found on hops from Blackman & Eastop (1984) was used to identify the aphid species. The key found in Cranshaw et al. (2018) was used to help distinguish between

alates of *P. humuli* and *P. cannabis*. Samples from throughout the season were stored in 70% ethanol and sent to Agriculture and Agri-Food Canada in Ottawa to confirm correct identification (Appendix A).

Identification of Natural Enemies

Natural enemies were identified to a functional group (i.e. predatory Diptera). In most cases, the digital microscope was not needed to identify natural enemies to this level. Samples of entomopathogenic fungi were collected and given to Agriculture Canada research station (Agassiz, B.C.). where they were plated out and sent to a federal laboratory in Ottawa for identification.

Analysis of Aphid Data Collected

Total aphids per field site, per week, and per sample in the field, were transcribed electronically from paper data sheets.

For each field site, information was recorded for each leaf, burr/cone or shoot. For every leaf, burr/cone or shoot taken the number of each aphid species was recorded. Of those aphid species found, the number that had wings and the number that did not have wings were then recorded.

For analysis of the aphid species, the total aphids found throughout the 10 sampling dates from shoots, leaves, burrs and cones from all sites were combined. The total number of each aphid species was calculated into a percentage of the total aphids found.

For analysis between aphid population dynamics between the two different hop varieties, only data on *P. humuli* were used. Weekly, the total number of *P. humuli* per field from leaf samples were tallied and divided by the number of leaves taken (50 per field site) to obtain the number of aphids per week for each field site.

Analysis of Natural Enemies Collected

For each field site, information was recorded for each leaf, burr/cone or shoot. For every leaf, burr/cone or shoot, the number of natural enemies, at each of its life stage, was recorded.

Predators and Parasitoids on Leaves

The total numbers of parasitoids and predators found on leaves from all field sites were broken down into aphidophagous life stages and non-aphidophagous life stages. The total number of all predators and parasitoids found on leaves from all field sites throughout the season was totalled and calculated into percentages per functional group (i.e. percent syrphid flies, percent ladybird beetles, etc.). The total number of aphidophagous stages of predators and parasitoids found on leaves from all field sites throughout the season was also totalled and calculated into percentages per functional group.

Entomopathogenic Fungus

Throughout the month of July, the total number of aphids killed by an entomopathogenic fungus was recorded per leaf per field. The percentage of aphid mortality as a result of this fungus was calculated by totalling all aphids found in the month of July per field site (including the aphids infected by the fungus) and dividing the number of aphids with the fugus by the total number of aphids found.

Predators and Parasitoids in Burrs and Cones

The total number of all predators found in burrs and cones from all field sites throughout the season was totalled and calculated into percentages per functional group.

2) Post-Harvest Field Study

Post-harvest, three of the six fields (Table 2) were monitored bi-weekly from September 21-November 16, 2019. On each sampling date, 25 leaves were taken from each field. Leaves were taken throughout the field in areas where hop vegetation remained alive. Leaves were removed from field and counted for aphids. Aphids were examined with the Dino-Lite Edge 1.3MP WF4115ZT for: 1) wing presence (alates); 2) wing absence (aptera); or 3) if wings were developing (altoids). The total count of aphids in each category was recorded per field and per week. For analysis, counts from each site per week were combined, and totals per category (winged, unwinged, and developing wings) were then calculated into percentages of totals.

3) Host Transfer Experiments

Two host transfer experiments were conducted outdoor at a 4 ha/10 acre fruit farm in Abbotsford, B.C. (Site A on Table 4; Fig. 6). The first experiment ran from June 2-July 14, 2019 and used alates (winged) of *P. humuli* (Experiment 1: Winged Transfer). The second experiment ran from July 29-August 26, 2019 and used aptera (unwinged) of *P. humuli* (Experiment 2: Unwinged Transfer). Experiment 1: Winged Transfer used hops, cannabis and nettle as treatments. Experiment 2: Unwinged Transfer used hops and two cultivars, or strains, of cannabis.

Design for Experiments 1 & 2

Both experiments consisted of four replicates of three treatments in a completely randomized design. Treatments differed between studies. In each experiment, Educational Science Giant Square Pop-Up Butterfly Cages (123 cm/48 inches x 67 cm/27 inches x 67 cm/27 inches) were used to contain individual plants so aphids could not escape, and so natural enemies could not get in (Fig. 5). Cages were arranged in two rows of six, and placed approximately 0.3 m/1 ft apart on a green tarp measuring 2.1 m/7 ft wide by 5.8 m/19 ft long (Fig. 5). The tarp and cages were placed on a south-facing slope (Fig. 5). The Set Seed function in R was used to randomly

assign plants to cages (Appendix B; Appendix D). Cages were labelled with flagging tape to indicate the plant type and replicate. Tents were weighted down with clean bricks and cleaned rocks.

Figure 5: Design for Host Transfer Experiments 1 & 2 with Educational Science Giant Square Pop-Up Butterfly Cages

Experiment 1: Winged Transfer

Alates (winged) of *P. humuli* reared on *Prunus* spp. were transferred to hops (Rosales: Cannabaceae), Cannabis (Rosales: Cannabaceae), and nettle (Rosales: Urticaceae) to assess the summer host range of the damson-hop aphid.

Aphid Rearing

Aphids were gathered early spring of 2019 from *Prunus* spp. to be used as rearing stock for the transfer study. *Phorodon humuli* aptera of were collected from two sites containing *Prunus* spp. from April 14-19, 2019.

Table 4: Description of *Phorodon humuli* collection sites in Abbotsford, B.C.

Collection site	Site Description	Plum type	Number of Plum trees
A	4 ha/10 acre organic apple orchard, Abbotsford, B.C.	Fruiting (yellow), cultivar unknown	30
B	Residential property in Abbotsford B.C. that neighbours Hop Site 5 of Field Survey	Ornamental (purple-type), cultivar unknown	2

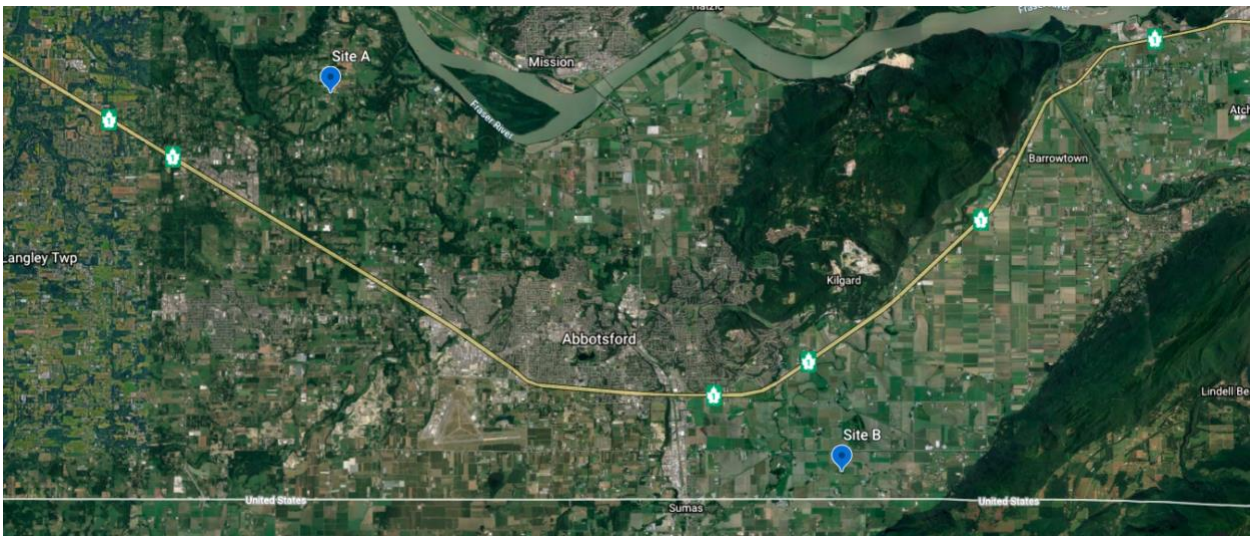


Figure 6: *Phorodon humuli* collection sites in Abbotsford, B.C. Experiments 1 & 2 performed at Site A

Aphid samples were sent to the Agriculture and Agri-Food Canada federal laboratory in Ottawa to confirm correct species for rearing (Appendix C). Leaves with aphids were collected in plastic containers with mesh lids. The collected aphids were put onto six potted plum suckers in four cages to exclude predators and were left to reproduce for six weeks (Fig. 7). Plum suckers were dug from plum Site A (Table 4) with permission of the property owner. Plum suckers were watered two times per week and leaves were checked weekly for aphid predators. Two sizes of mesh cages were used. The large rearing cages measured 72 in/183 cm high x 23 in/58 cm deep x 23 in/58 cm wide. The smaller cages measured 48 in/123 cm high x 27 in/67 cm deep x 27 in/67 cm wide.

Figure 7: Mesh cages containing plum suckers used to rear *Phorodon humuli* at Site A

Treatments and Corresponding Materials

Hop, nettle and cannabis plants were the three treatments used. Propagated hop plants, *Humulus lupulus,* from vegetative cuttings were purchased from Green Flora Nursery in Abbotsford, B.C.. The hop cultivar used was 'Cascade.' Propagated cannabis plants, *Cannabis sativa,* from vegetative cutting were donated from a commercial producer in the Fraser Valley. The cultivar, or strain, used was 'Afghan Kush.' Nettle, *Urtica dioica,* was foraged and dug up from Site A (Table 4; Fig. 6).

Hops, cannabis and nettle were transplanted into 15 L/5 gallon aeration fabric pots (247 Garden 5 Gallon Grow Bags/Aeration Fabric Pots w/Handles) on May 19, 2019. Pot diameters measured 30 cm/12 inches and pot height measured 25 cm/10 inches (Fig. 9). Pots were filled with approximately 14.1 L/12.8 dry quarts of Fox Farm Ocean Forest Potting Soil.

Plants were not pruned throughout the course of this study. Plants were watered as needed throughout the study (two to four times a week). Plants were not fed any nutrients outside of the initial nutrients in the potting mix.

Data Collection and Analysis

Each plant was placed into the aphid rearing cages (Fig. 7) for a 24-hour period to be colonized by aphids. This allowed for alates of *P. humuli* to be transferred to the plant without any handling which could have resulted in damage to the aphids. After 24 hours, the number of aphids on each plant was recorded, and placed into its assigned, empty cage. Aphid numbers

were then recorded weekly for five weeks from June 2 – July 14, 2019. During each recording period, plants were removed briefly from the cage and leaves were examined for aphids.
Initial starting population levels of winged aphids were different among treatments at the start of the trial, thus data was analysed using a one-way ANOVA examining effect of host plant on the percent change in counts between week 0 and week 5.

Experiment 2: Unwinged Transfer Experiment

Apterous *P. humuli* reared on hops were transferred to hops and two cultivars of cannabis to assess the suitability of cannabis as a summer-host of hop aphid.

Aphid Rearing

Hop plants with high levels of *P. humuli* from Experiment 1: Winged Transfer were kept in their original cage after the experiment had finished, and were used to inoculate plants in Experiment 2: Unwinged Transfer.

Treatments and Corresponding Materials

Hops and two cultivars of cannabis were used as the three treatments Experiment 2: Unwinged Transfer. Propagated hop plants from vegetative cuttings were purchased from Green Flora Nursery in the Fraser Valley, B.C.. The hop cultivar, 'Cascade' was used again in Experiment 2. Two cultivars, or strains, of cannabis propagated from vegetative cuttings were donated from a commercial producer in the Fraser Valley: 'Afghan Kush' and 'Strawberry Switchblade.' Plants were potted into 15 L/5 gallon aeration fabric pots (247 Garden 5 Gallon Grow Bags/Aeration Fabric Pots w/Handles) (Fig. 8). Pot diameters measured 30 cm/12 inches and pot height measured 25 cm/12 inches. Pots were filled with approximately 14.1 L/12.8 dry quarts of Fox Farm Ocean Forest Potting Soil.

Plants were not pruned throughout the course of this study. Plants were watered three-five times a week, depending on the weather. Plants were not fed any nutrients outside of the initial nutrients in the potting mix.

Data Collection and Analysis

Aptera of *P. humuli* from hop plants in the Winged Transfer Study were used to inoculate plants in the Unwinged Transfer Study. Two leaves with aphids were removed from the infested hop plants. Aphids on the two leaves were counted and recorded, then placed onto clean plants in cages (Fig. 9). Each plant received 200 aphids by this method (Fig. 9). Aphid numbers were recorded weekly for a total of four weeks from July 29-August 26, 2019.
Data was analysed using repeated measures MANOVA. All data were analysed in JMP – version 5.1 (SAS Institute, Chicago, IL).

Figure 8: ‘Strawberry switchblade’ in 15 L/5 gallon fabric pot

Figure 9: Aphid inoculation method used in Experiment 2: Unwinged Transfer

PROJECT RESULTS

Pre-Harvest Field Survey: Aphids

Aphid Species

The first objective in this study was to determine aphid species composition in Fraser Valley hop fields. On all hop plant materials monitored, 17,369 out of 17,653 aphids found throughout the season were *P. humuli* which accounts for 99% of all aphids found (Fig. 10). All six sites had *P. humuli* (Table 5). Of the other species found in fields besides *P. humuli*, 227 were *Aphis fabae*, commonly known as black bean aphid and 53 were *Macrosiphum euphorbiae*, known as the potato aphid and four were ‘incidental’ aphids (Fig. 10). Of these four aphids, two aphids were *Brachycaudus helichrysi* (Kaltenbach) (Fig. 10), also commonly known as the leaf curl plum aphid and leaf-curling plum aphid; and two were unknown. The two unknown aphids were also sent for identification, but were not identified. The other aphid species including *A. fabae, M. euphorbiae,* and incidentals made up 1% of all aphids found (Fig. 10) and were not present in all fields (Table 5).

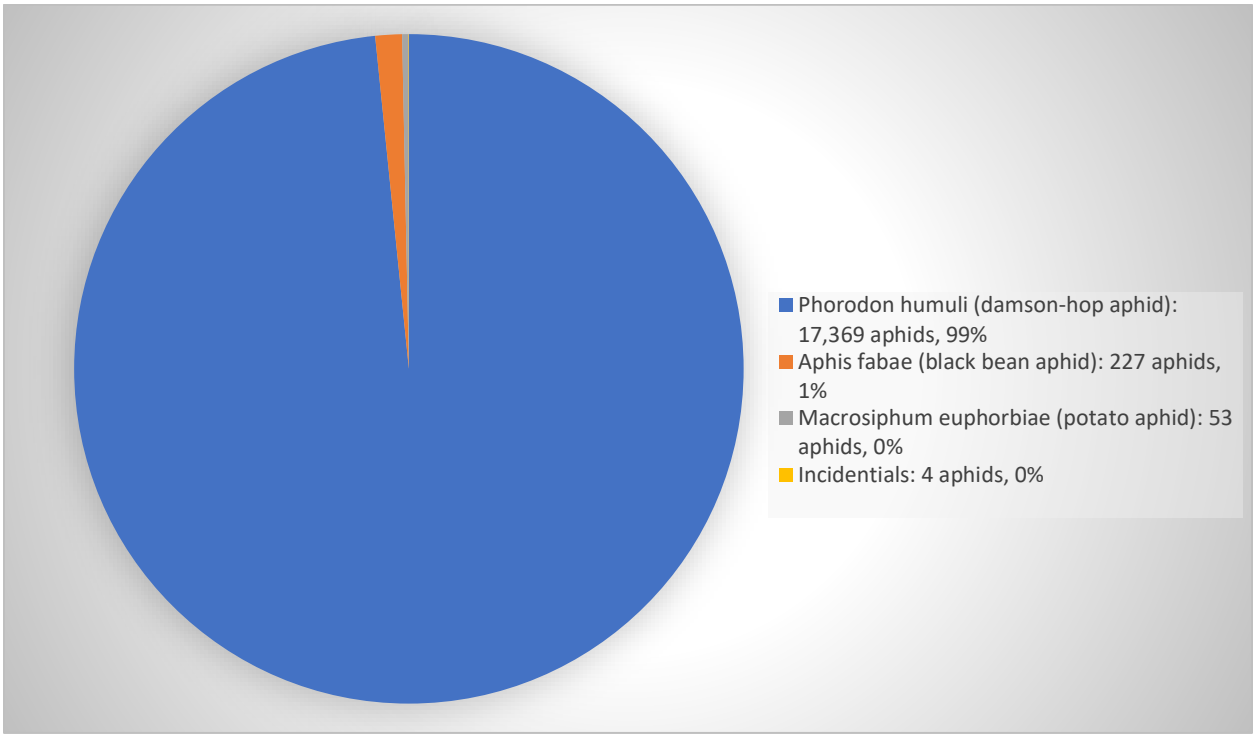


Figure 10: Composition of aphid species found in six commercial hop yards in the Fraser Valley, B.C. from May 10 – September 13, 2019. Total aphids = 17,653 on hop shoots, leaves, burrs and cones.

Table 5: Aphid species present by site in six hop fields in Fraser Valley, B.C. in 2019

		Aphid species present				Description of adjacent land surrounding farm									
Farm /site	Hop cultivar used in study	*Phorodon humuli* (damson-hop aphid)	*Aphis fabae* (black bean aphid)	*Macrosiphum euphorbiae* (potato aphid)	Incidentals	Hay	Berries	Corn	Poultry	Mixed flower	Mixed veg	Hemp	Forest	Riparian	Golf course
1	Cascade	X	X	X	X	X	X			X			X	X	
2	Triple Pearle	X	X				X	X						X	
3	Triple Pearle	X	X	X	X			X	X	X					X
4	Cascade	X		X	X		X					X			X
5	Triple Pearle	X				X		X			X				
6	Cascade	X					X		X						X

Phenology Aphids in Hop Fields

Phorodon humuli was absent in fields in the first week (May 10, 2019). The first date *P. humuli* was recorded in hopyards was May 24, 2019 and were found weekly until harvest. From May 24-July 5, 2019, alates of *P. humuli* were present in hopyards weekly. No alates were seen in hop fields after July 5 until after harvest. From July 19 onwards, *P. humuli* was the only aphid

species found in the six hopyards. *Phorodon humuli* was the only aphid species found in cones or burrs, however, the incidence of this was low, and only in fields with high levels of aphids.

Aphid species other than *P. humuli* were found in hopyards from May 10 – July 5, 2019. *Aphis fabae* and *M. euphorbiae* were the first aphids to be found in hopyards, on May 10, 2019. Aphid species other than *P. humuli* were found early in the season, but were not found after July 5, 2019.

Phenology of *Phorodon humuli* by Hop Cultivar

Levels of spring migrants of *P. humuli* started low in both 'Cascade' and 'Triple Pearle' fields (Figs. 11 & 12). In 'Triple Pearle' fields, peak population of *P. humuli* was the week of June 21 (Fig. 12). In all three Triple Pearle sites, one application of flupyradifurone (Table 6) brought levels below the provisional threshold of 8-10 aphids per leaf (Fig. 12). At Site 6, no spray was required (Table 6). At Site 1, insecticidal soap was used (Table 6) and aphid levels were not brought below the provisional threshold (Fig. 11).

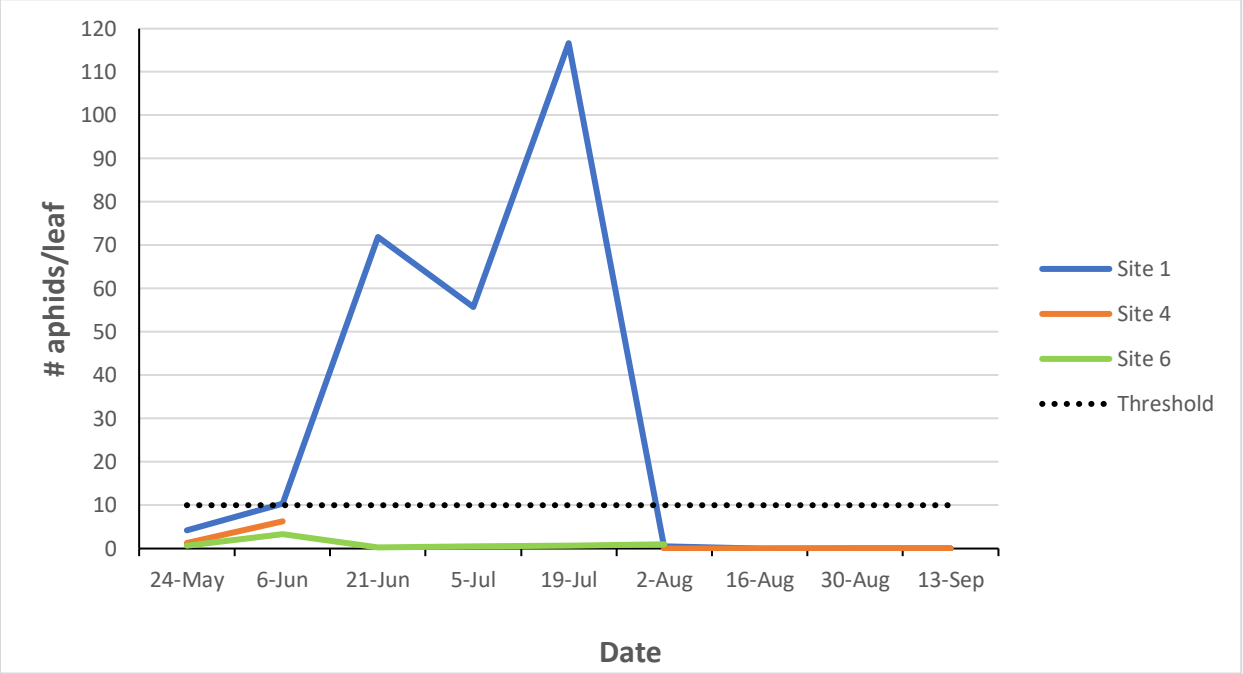


Figure 11: Number of *Phorodon humuli*/leaf/week over nine weeks in three 'Cascade' fields (Site 1, Site 4 and Site 6) relative to a provisional action threshold of 10 aphids/leaf

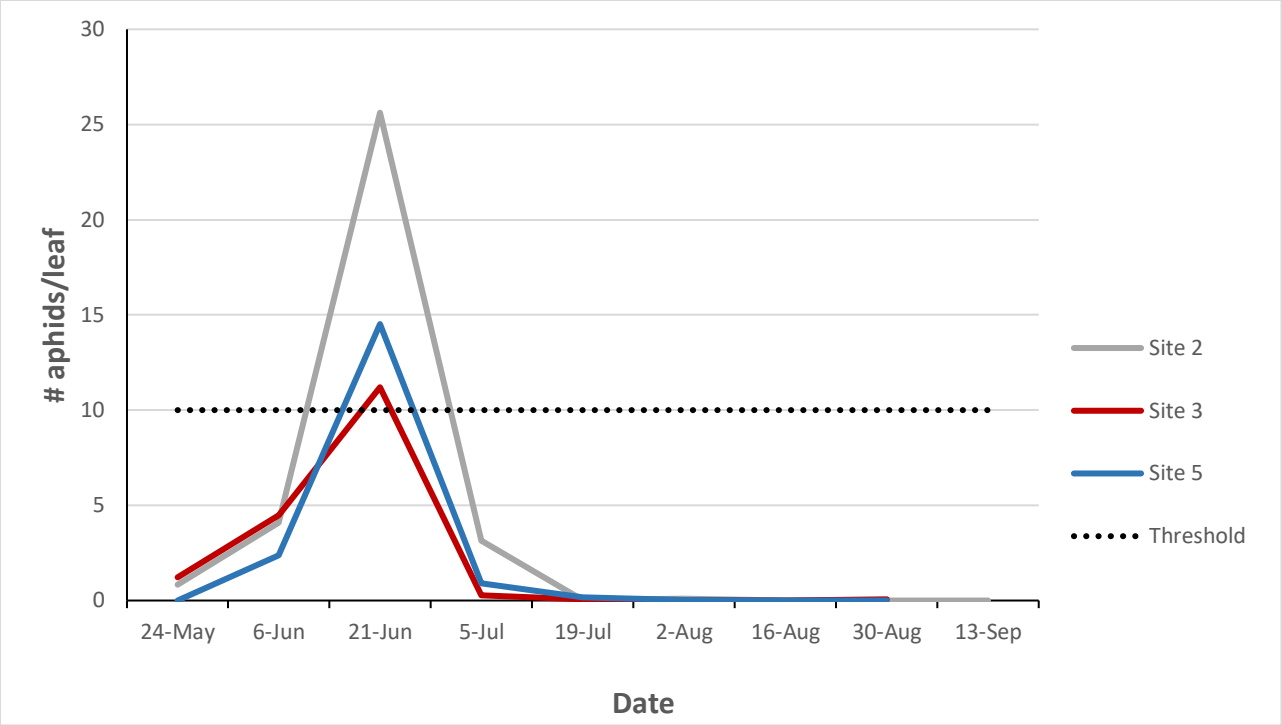


Figure 12: Number of *Phorodon humuli*/leaf/week over nine weeks in three 'Triple Pearle' fields (Site 2, Site 3 and Site 5) relative to a provisional action threshold of 10 aphids/leaf

Table 6: Insecticides used to target *Phorodon humuli* at six Fraser Valley sites during 2019

Farm/Site	**Date of application(s)**	**# of applications**	**Active Used**	**Tradename**
1	July 4, July 24, Aug.3	3	Potassium salts of fatty acids	Kopa
2	July 3	1	Flupyradifurone	Sivanto
3	June 25	1	Flupyradifurone	Sivanto
4	June 28	1	Flupyradifurone	Sivanto
5	July 4	1	Flupyradifurone	Sivanto
6	N/A	0	N/A	N/A

With the exception of Site 1 (cultivar 'Cascade'), which had high levels of aphids in part due to the inefficacy of insecticidal soap, 'Triple Pearle' fields had higher levels of *P. humuli* compared to 'Cascade' (Figs. 11 & 12).

Pre-Harvest Field Study: Natural Enemies

Predators

The second objective was to determine the associated aphidophagous natural enemies in hop fields the Fraser Valley, British Columbia. Aphid predators were the most abundant group of natural enemies compared to parasitoids or entomopathic fungi. Ladybird beetles, syrphid flies, lacewings, aphid midges, and minute pirate bugs were found in hopyards throughout the season (Fig. 13). Ladybird beetles were the most abundant aphid predator found on leaves in hopyards, contributing to 62% of the predators found (Fig. 13). Syrphids were next most abundant predator found, making up 16% of the predators seen (Fig. 13). Lacewings were the third most common predator at 13% of the total predators found (Fig. 13).

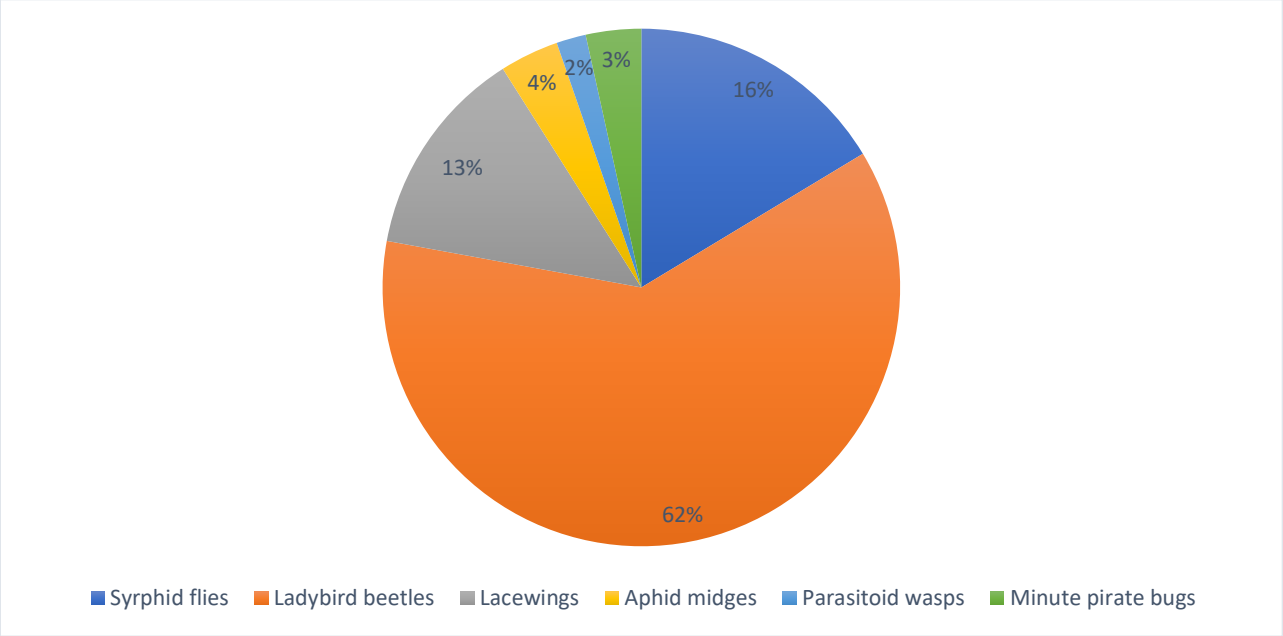


Figure 13: Total predators and parasitoids (all life stages) from all sites found on leaves from May 24 – September 13, 2019. Total leaves = 2350; total predators = 434

When non-aphidophagous stages (i.e. eggs) were removed, ladybirds were still most abundant, representing 76% of all predators (Fig. 14). Syrphids were still second most abundant, representing 11% of all predators (Fig. 14). Lacewings, however, only represented 1% of predators when non-aphidophagous stages were removed (Fig. 14). The majority of lacewings found were eggs (Fig. 15).

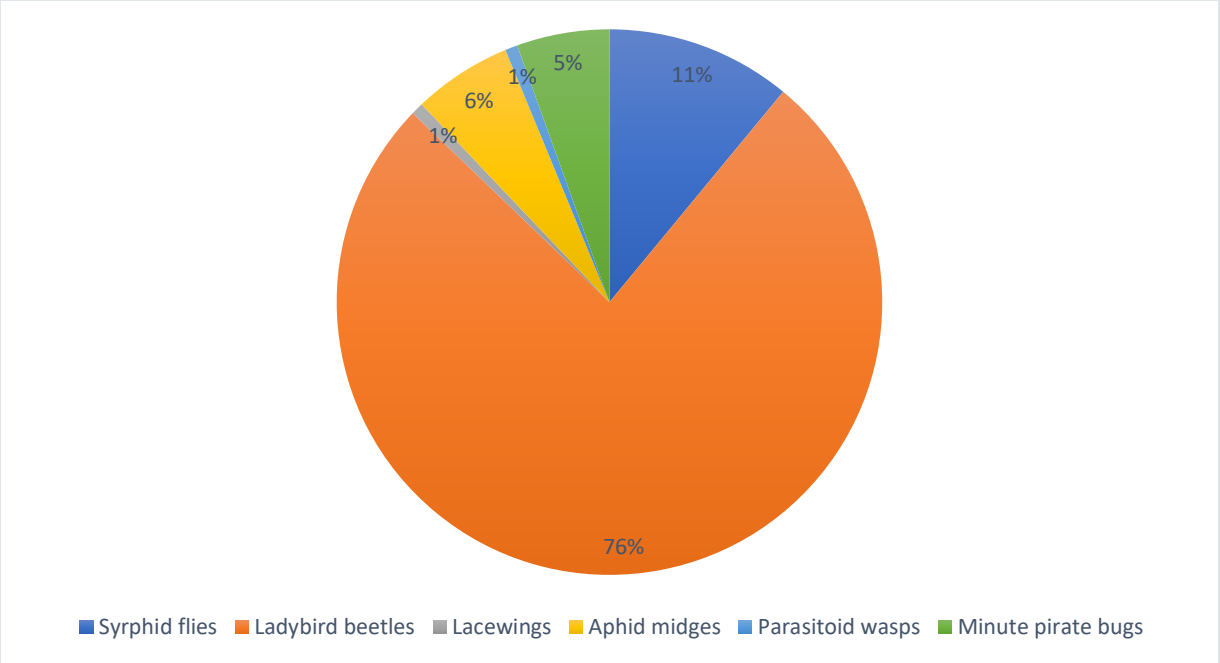


Figure 14: Total aphidophagous immatures and adults from all sites (found on leaves) from May 24 – September 13, 2019, on 2350 leaves. Total aphidophagous immatures and adults = 273

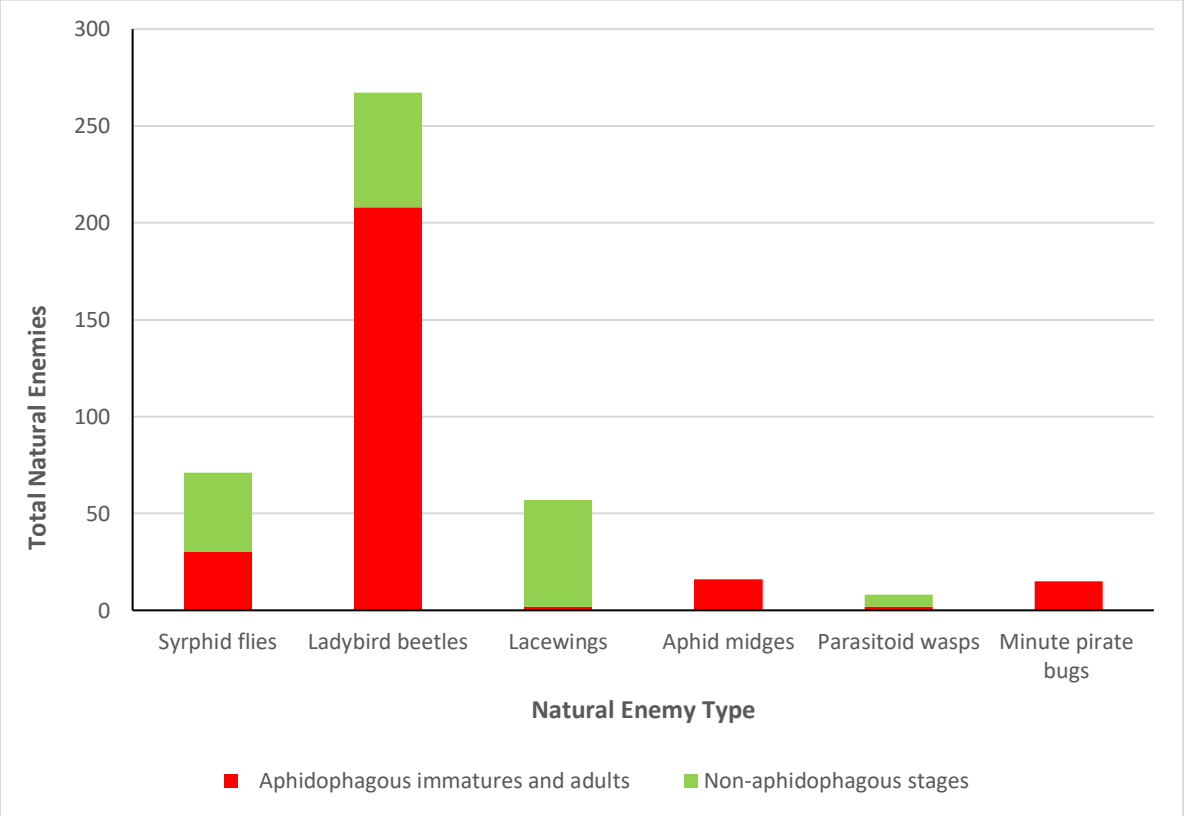


Figure 15: Comparison of aphidophagous stages to non-aphidophagous stages found leaves from all sites from May 24 – September 13, 2019. Total leaves = 2350; total predators = 434

Only six aphid mummies (parasitized aphids) were found during the ten sampling dates from May 10-Sept. 13, 2019 (Figs. 14 & 15). Sixteen aphid midges (*Aphidoletes* spp.) were found (Figs. 13-15). Fifteen minute pirate bugs (*Orius* spp.) were found through leaf sampling (Figs. 13-15).

Predators in Cones

In burrs/cones from July 19–September 13, 2019, 27 predators were found. *Orius* spp. and ladybird beetle larvae were the only predators found in cones (Fig. 16). Of the 27 predators, 25 were *Orius* spp. which represents 93% of predators found in cones (Fig. 16). Of the 25 *Orius* spp. found in burrs/cones (Fig. 16), 16 were nymphs and 9 were adults. Only two ladybird beetles were found (Fig. 16) and both were larvae.

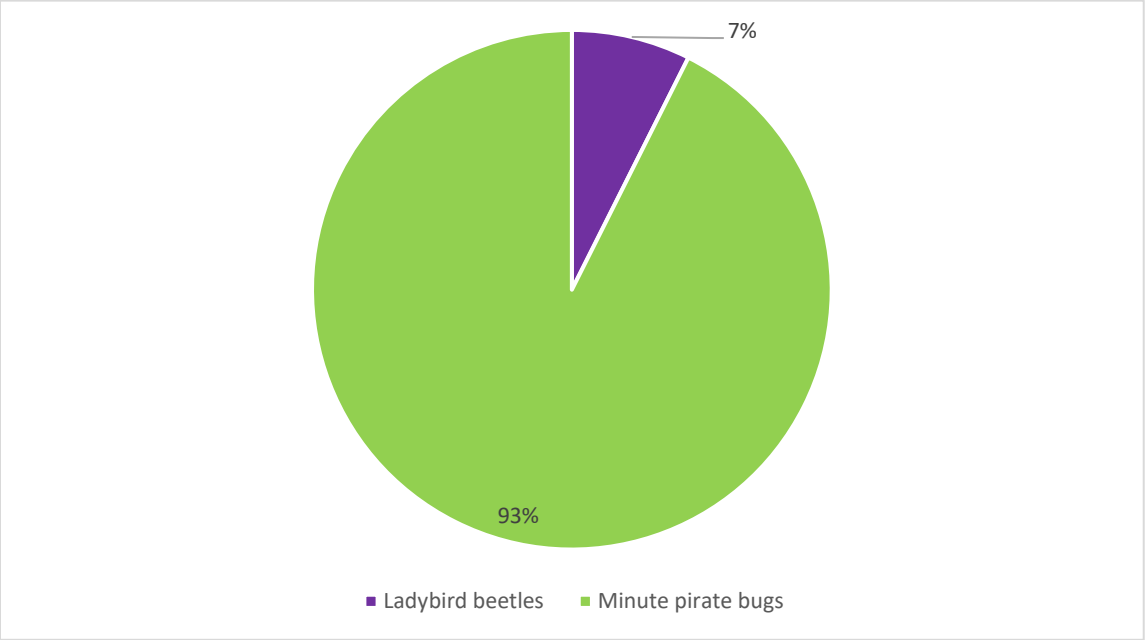
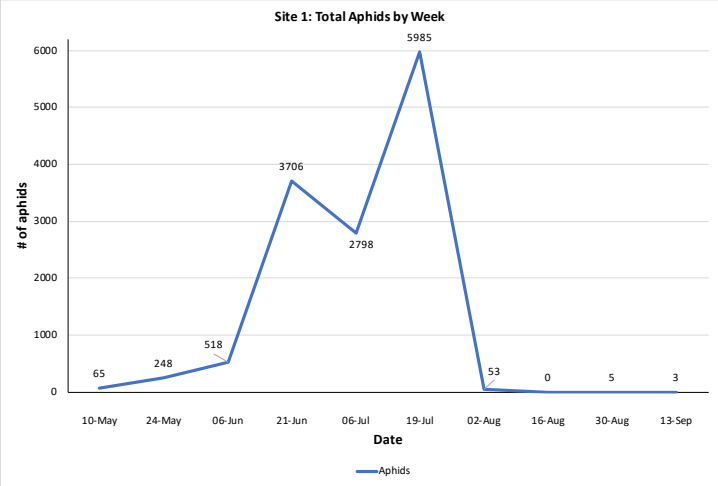
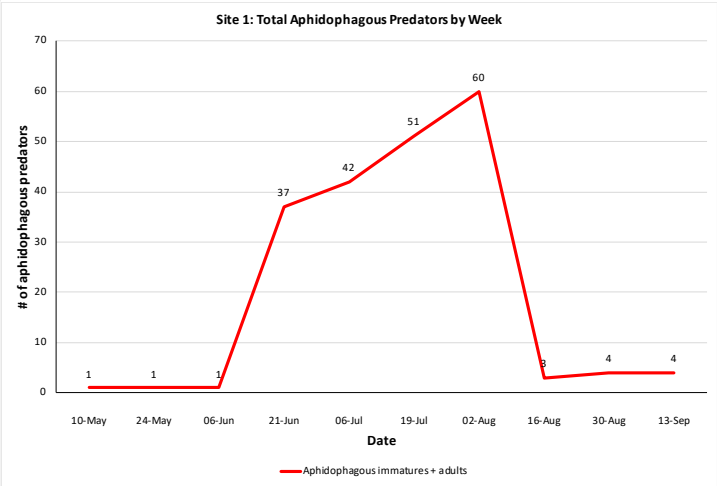


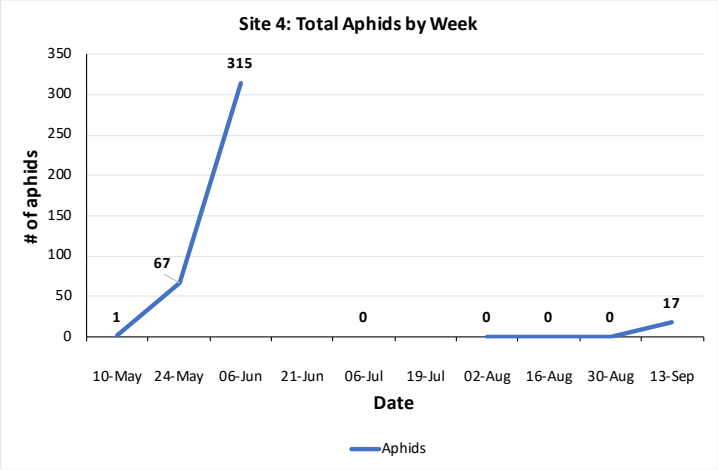
Figure 16: Total predators in burrs/cones from July 19 – September 13, 2019. Total cones/burrs = 928. Total predators = 27

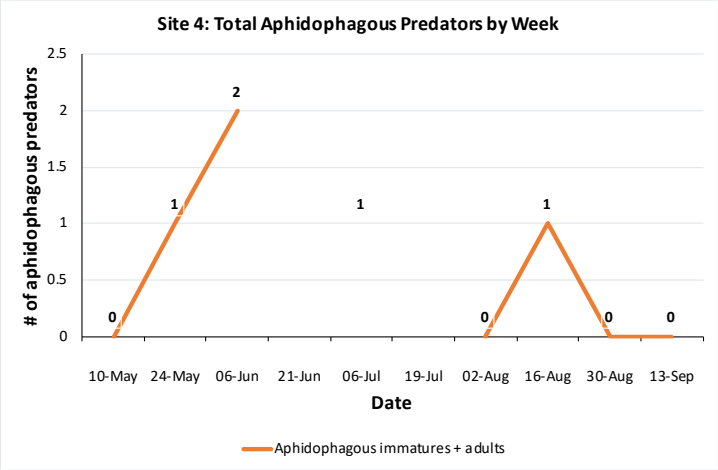
Total Predators per Week vs. Total Aphids per Week

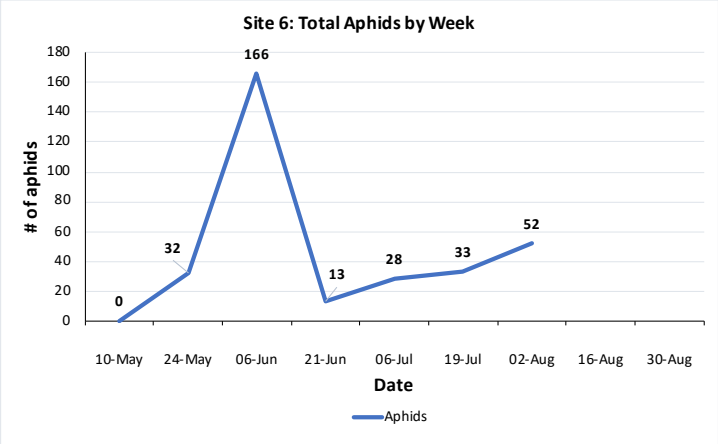
At all six sites, abundance of predators was low compared to aphids (Figs. 17 & 18). Similarly, predator population growth was low compared to aphid population growth in both 'Cascade' fields (Fig. 17) and 'Triple Pearle' fields (Fig. 18).











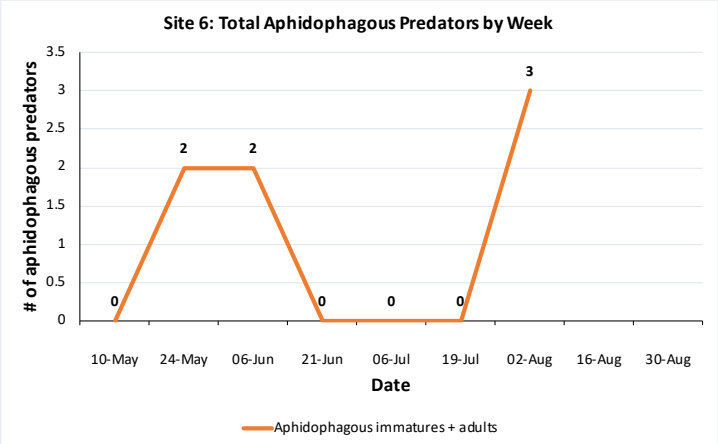


Figure 17: 2019 total aphids (all species, winged and unwinged) combined on all parts of plants per week compared to total predators on all parts of plants per week at ‘Cascade’ sites

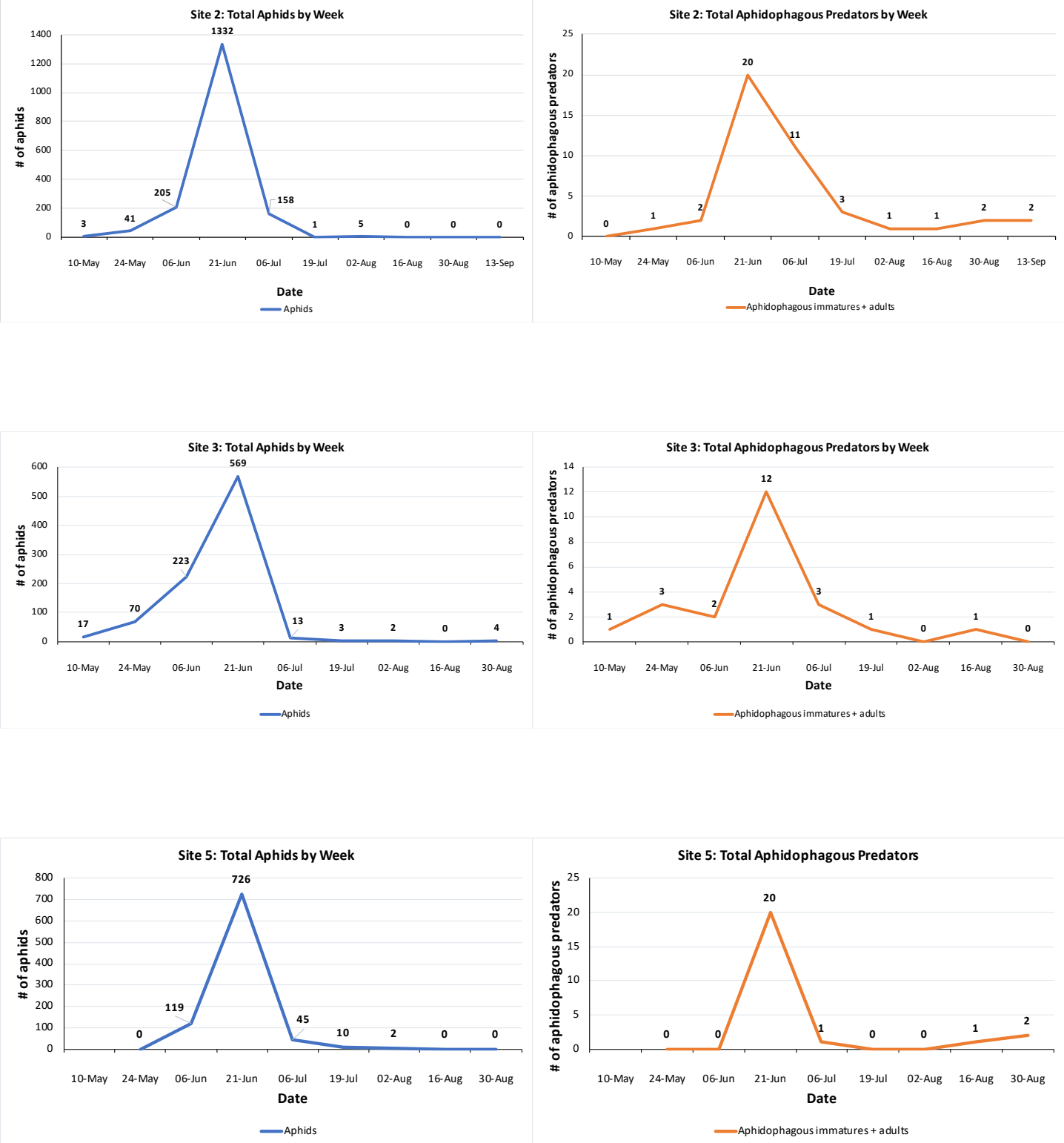


Figure 18: 2019 total aphids (all species, winged and unwinged) combined on all parts of plants per week compared to total predators on all parts of plants per week at ‘Triple Pearle’ sites

Entomopathogenic Fungus

An entomopathogenic fungus was found during the month of July (July 5 – July 19, 2019) (Fig. 19). This fungus was present at four of the six sites. Aphids infected by the fungus could be distinguished from healthy *P. humuli* due to the unusual peach colour. Of those four sites where the fungus was present during the month of July, it contributed to 12% aphid mortality at Site 1; 25% aphid mortality at Site 2; 0.04% at Site 5; and 0.02% at Site 6. Of those aphids killed by this fungus, all were unwinged except for one. The fungus was identified as *Cladosporium* spp. by Agriculture and Agri-Foods Canada (Appendix E).

Figure 19: *Phorodon humuli* killed by an entomopathogenic fungus during month of July, 2019

Post-Harvest Field Survey: Aphids

No alates of *P. humuli* were found in the six hop fields prior to harvest. The first alate to be seen in the fall was on September 21, 2019. Unwinged, winged and aphids developing wings were found after harvest from September 21 – November 3, 2019 (Table 7). The aphids were found on unharvested plants, around structural posts, in the regrowth, and on any hop vegetation that was still alive.

Table 7: Post-harvest, bi-weekly combined averages of *Phorodon humuli* from three sites from September 21 – November 16, 2019

Date	Average # *Phorodon humuli* per leaf	Percent unwinged	Percent developing wings (wings not fully formed)	Percent winged (fully developed)
September 21	0.8	98.33%	0.00%	1.67%
October 6	0.52	61.54%	34.62%	3.85%
October 19	1.54	51.95%	44.16%	3.90%
November 3	0.78	50.85%	49.15%	0.00%
November 16	0.0	0.00%	0.00%	0.00%

Host Transfer Experiments

The third objective was to determine if *P. humuli* had secondary summer hosts other than hops. This was accomplished by two different experiments. Experiment 1: Winged Transfer, involved transferring winged *P. humuli* from *Prunus* spp. to hops, cannabis and nettle. Experiment 2: Unwinged Transfer, involved transferring unwinged *P. humuli* from hops to hops and two different cultivars, or strains, of cannabis (Unwinged Transfer Study).

Experiment 1: Winged Transfer

Transferring winged aphids from *Prunus* spp. to different hosts proved to be challenging (Appendix F). The initial number of aphids surviving transfer at the start of the trial was significantly different across the transfer host species ($F(2,9)=9.44$, $p=0.006$). Thus, the data at the end of the trial were analyzed to examine the population change from the initial counts at the start of the trial to the final counts at Week 5. The only transfer host that hop aphid populations increased on was hops. For nettle and cannabis the populations declined leading to significant treatment differences between hops and cannabis and hops and nettle (Fig. 20; $F(2,9)=13.09$, $p=0.002$). Nettle and Cannabis were not significantly different from each other based on Tukey Kramer HSD (Fig. 20).

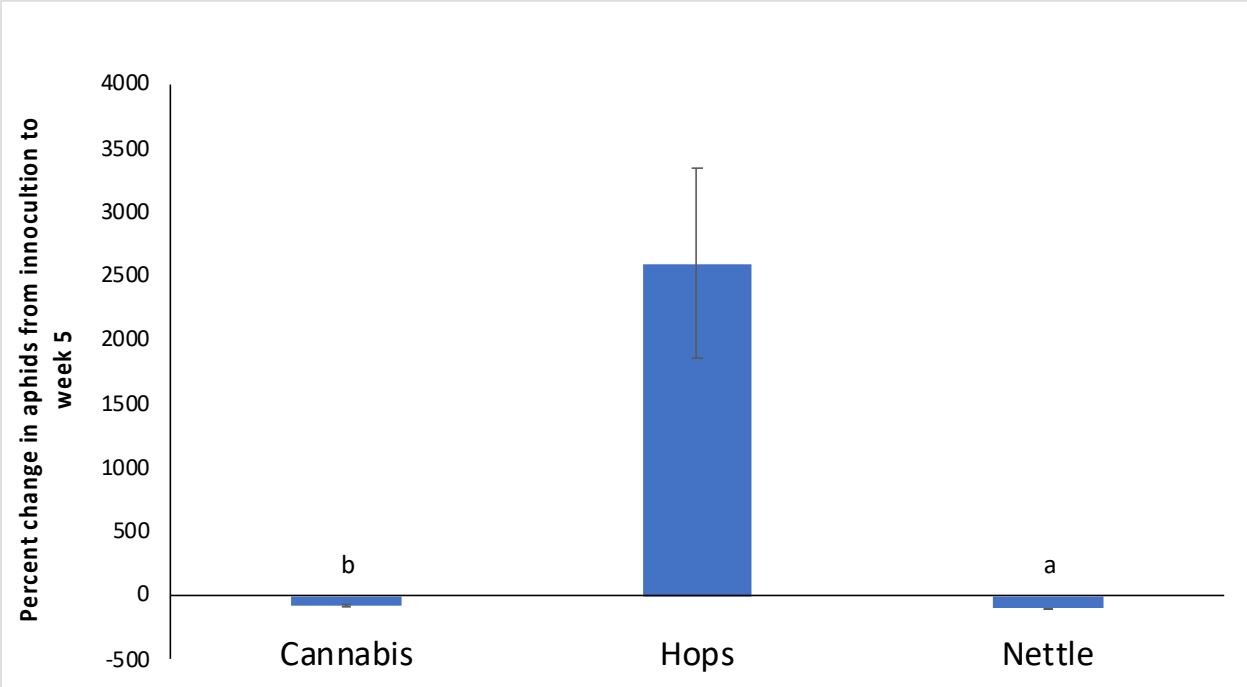


Figure 20: Effect of host plant on the percent change in aphid counts between week 0 (inoculation) and week 5. $F(2,9)=13.09$, $p=0.002$.

Experiment 2: Unwinged Transfer

Over the course of 4 weeks, aphid decline in both cannabis treatments and levels increased on hops (Fig. 21). There were significant differences in the survival of aphid populations on the three different transfer hosts during the last two weeks of the four week study, but not the first two weeks (Fig. 21; Treatment: $F(2,9)=6.21$, $p=0.02$; Time: $F(4,6)=45.99$, $p=0.0001$; Treatment X Time: Wilks Lambda=0.03, $F(8,12)=6.68$, $p=0.002$).

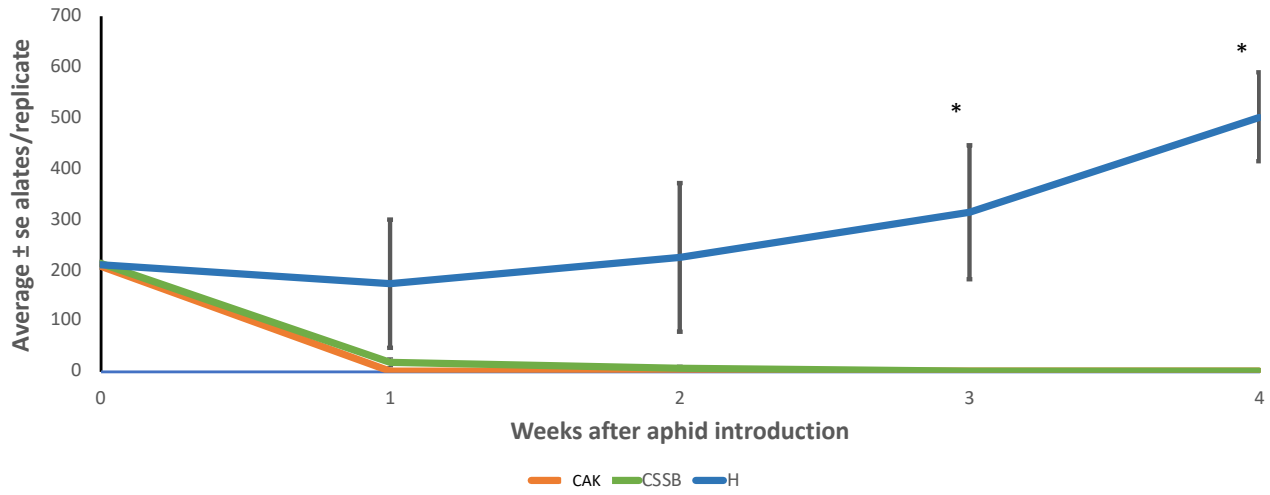


Figure 21: Survival of wingless damson-hop aphids (*Phorodon humuli*) on three different hosts: *Cannabis sativa* 'Afghan Kush'= (CAK); *Cannabis sativa* 'Strawberry Switchblade' = (CSSB); and *Humulus lupus* 'Cascade'= (H) over the course of 4 weeks. Data points are the mean (±s.e.) number of wingless aphids on a plant (replicate), with four replicates per treatment/week. Asterisks indicate significant differences between treatments for the specific week.

DISCUSSION

Field Survey Results: Aphid Species in Fraser Valley Hop Fields

The first objective in this study was to determine aphid species composition in Fraser Valley hop fields.

Phorodon humuli

Phorodon humuli was the dominant aphid species found in all six fields in 2019 (Fig. 10). Population levels started low in the spring in Fraser Valley hop fields. The low numbers of spring migrants in the Fraser Valley could be attributed to the lack of abundant overwintering sources. The presence and amount of both *Prunus* spp. and hops has been shown to influence *P. humuli* abundance in both Washington (Wright et al., 1995) and England (Taylor et al., 1979). In areas containing more *Prunus* spp. hosts compared to hopyards, such as areas of England and Vancouver WA, more spring migrants of *P .humuli* have been recorded (Taylor et al., 1979; Wright et al., 1995). In areas with more hopyards and less *Prunus*, there were higher recording of fall migrants (Wright et al., 1995). Similarly, in Prosser, WA, Campbell and Cone (1994) reported very few spring migrants, and high numbers of migrants in the fall, and attribute this to the small amounts of appropriate winter hosts in the area.

The development of *P. humuli* alates in the fall occurred after harvest in Fraser Valley hop fields (Table 7). Over the course a 10 year study in Washington, the first winged gynoparae were found

from August 27 – September 12, which corresponded with an average of 1,244 accumulated degree-days (Wright et al., 1995). Wright et al. (1995) describe that in the fall, winged females are produced prior to males. In Washington, winged females are first seen in late August, and males in mid-September (Wright et al., 2005). In the Fraser Valley hop fields, the first alate to be seen in the fall was on September 21, 2019, which is later than in Washington.

Unwinged, winged and aphids developing wings were found on leaves of unharvested hops and hop regrowth in hop fields after harvest until November 3, 2019, ranging from 0.5-1.54 aphids/leaf (Table 7). This finding is similar to Wright et al. (2005), who found that remaining hop material after harvest supported levels of *P. humuli* ranging from 1.7-5.8/leaf. After November 3, 2019, all foliage was frost killed in the three Fraser Valley hop fields. Wright et al. (1995) linked the end of fall migration with the killing of hop foliage by frost.

'Cascade' has been noted for its susceptibility to aphids (Dorschner & Baird, 1988). In this field study, the incidence of higher number of aphids in 'Triple Pearle' may be a result of the growth habit of the plant. 'Triple Pearle' has a vigorous growth and has a dense canopy. Hop aphids develop quickly in cooler weather (Calderwood et al., 2015; Morrison, 1958). The thick canopy of 'Triple Pearle' may provide some protection from the direct sunlight and provide a microclimate more suitable for *P. humuli* in the Fraser Valley.

Other Aphid Species

Other aphids than *P. humuli* made up only 1% of all aphids found in the surveyed Fraser Valley hop fields (Fig. 10). No cannabis aphids (*P. cannabis*) were found, supporting Cranshaw et al.'s (2018) statement that this aphid is monophagous on cannabis/hemp.

The surrounding crops can aid in explaining the presence of aphid species other than *P. humuli* in Fraser Valley hopyards. Of the identified incidentals (Fig. 10), *B. helichrysi*, was found at Site 3 (Table 5). This aphid is an important aphid on sunflowers (Lerin & Badenhausser, 1995). Site 3 is in close proximity to mixed flower production (Table 5) and hosts an annual sunflower festival.

Aphis fabae and *M. euphorbiae* have large host ranges, and Blackman and Eastop (1984) included both these species as aphids on hops, so it is not surprising they would be found in the Fraser Valley hop fields. The presence of these species was confined to spring and early summer. The presence of both weeds and cultural practices may have impacted the timing of when these aphids were present in hop fields. All six sites in this study had high weed pressure. While *A. fabae* was found on hops (Fig. 22), it was also commonly observed on weeds in close proximity to the hop plants (Fig. 23). Black bean aphid was found on *Galium aparine*, often referred to as cleavers; *Chenopodium album* (lambsquater); and *Rumex crispus* (curly doc). Soon after weeds were controlled (Table 8), neither *A. fabae* nor *M. euphorbiae* were found in fields. Typically weed control and deleafing of the lower hop leaves (to promote air flow to reduce downy mildew) are done simultaneously with the same herbicide product. The timing of this weed control and deleafing in the Fraser Valley hop fields removed both weed hosts and lower hop leaves (Table 8) removing all suitable food sources for aphids.

Figure 22: *Aphis fabae* (black bean aphids) on hops

Figure 23: *Aphis fabae* (black bean aphids) on weeds (cleaver) in hopyard

Table 8: 2019 herbicide summary application for six Fraser Valley fields

Farm/Site	Date of application(s)	# of applications	Active Used	Tradename
1	July 17 & 27	2	Carfentrazone-ethyl	Aim
2	July 19	1	Glufosinate ammonium	Ignite
3	June 27; Aug. 6	2	Glufosinate ammonium	Ignite
4	June 21; July 19	1	Carfentrazone-ethyl	Aim
5	N/A	0	N/A	N/A
6	June 18	1	Carfentrazone-ethyl	Aim

While other species were present in hop fields, they were not present after deleafing or after pesticide application to control *P. humuli.* Other aphids than *P. humuli* made up only 1% of all aphids found (Fig. 10) and were not found in the cones. They were not found at all six sites (Table 5). For those listed reasons, they are not of great concern in the surveyed Fraser Valley hopyards.

Field Survey Results: Natural Enemies

The second objective was to determine the associated aphidophagous natural enemies in hop fields the Fraser Valley, British Columbia.

Predators

Two main aphidophagous arthropod families that have received attention for their role in aphid management in hop yards are Coccinellidae (ladybird beetles) and Anthocoridae (minute pirate bugs or flower bugs). Ladybird beetles were the most abundant predators on leaves in this Fraser Valley survey (Figs. 13 & 14), while minute pirate bugs were the main predator found in cones (Fig. 16). Ladybird beetle abundance in this 2019 Fraser Valley survey is similar to the findings in Washington by Campbell and Cone (1994) and in Oregon by Woods et. al (2014). Similar to the findings of Lorenzana et al. (2010), *Orius* spp. was the most common predator found in cones. In this 2019 Fraser Valley study, *Orius* spp. was not commonly found on hop leaves (Figs. 13 & 14) however, this could be explained by the leaf sampling method used in the study. Campbell (1978) argued that leaf sampling underestimates anthocorids because they often hide between the trellis strings and hop stems.

Syprhid flies were the second most common predator in hop fields (Figs. 13-15).This finding differs from that in Washington, where Campbell & Cone (1994) where predatory Diptera (including syrphids) were uncommon, and in England, where syprhids were rarely seen (Barber et al., 2003). It is however, in alignment with Woods et al. (2004), who found that syrphids were present in Oregon hop fields, but in low numbers. While syrphid flies were the second most abundant predator, they only made up 11% of all aphidophagous predators found in this study (Fig. 14).This finding is also similar to Campbell (1978) who noted that syprhids were present by the end of June, but not in high levels. Likewise, it is similar to Aveling (1981), who noted that two syrphid species were found in English hopyards, *Syrphus ribesii* and *S. balteatus*, however, were found in low numbers in comparison to other predators. In post-harvest checks, syrphids were the only predator found. This corresponds with James & Dreves (2015), who have observed that syrphids overwintering as pupae in hopyards.

Aphidoletes spp. (aphid midges) made up 6% of all aphidophagous predators found (Fig. 14). Woods et al. (2014) did not note any Cecidomyiidae in their study, however *Aphidoletes* spp., has been known to occur in other Oregon fields (James & Dreves, 2015). Like Campbell & Cone (1994), Cecidomyiidae were not found regularly or abundantly in Washington hop fields. In the two-year study by Campbell (1978), larvae of *Aphidoletes* spp. were only found in one year in the untreated plots where *P. humuli* levels were high.

The majority of lacewings found in this study were eggs (Fig. 15). Aphidophagous stages of lacewings made up only 1% of all predators found (Fig. 14). Campbell & Cone (1994) note that while many lacewings were found in their Washington study, the eggs were frequently parasitized by *Telenomus* spp. Intraguild predation may have contributed to the low incidence of other stages such as larvae or adults in this 2019 Fraser Valley field study, however, *Telenomus* spp. were not observed. Lacewing larvae are also often nocturnal (Van der Ent et al., 2017) and

sampling in this study took place during the day. This may be another reason as to why numbers of aphidophagous stages were low.

Parasitoids

In this 2019 Fraser Valley field survey, aphid parasitoids and aphid mummies were rarely found in hop fields (Figs. 13-15). This is similar to the reporting of parasitoids in Washington (Campbell & Cone, 1994; Pike & Starý, 1995). Campbell's 1978 reporting of less than 1% parasitism are on par with this study, confirming that aphid parasitoids currently do not play a significant role in naturally occurring biological controls in hop fields in the Fraser Valley.

While aphid mummies and adult parasitoids were not commonly found in hopyards, during the collection of *P. humuli* from *Prunus* spp. for rearing purposes for the two transfer studies, parasitoids were seen, but not recorded (Fig. 24). The rarity of parasitization on hops compared to the more frequent parasitization on *Prunus* spp. corresponds with the findings of Wright & James (2001). There have been several suggestions as to why there are so few aphid parasitoids of *P. humuli* while on hops, and all of which are compelling arguments to explain the findings of this 2019 Fraser Valley field survey. *P. humuli* originates from the West-Palearctic (comprised of Europe, North Africa, northern and central parts of the Arabian Peninsula, and part of temperate Asia) (Pike & Starý, 1995). Wright & James (2001) and Pike & Starý (1995) have suggested that native parasitoids have not yet been able to adapt to *P. humuli* because it is an introduced species to North America. Pike & Starý (1995) have also suggested that the use of pesticides in commercial fields can help explain the low numbers of parasitoids in hop fields. Copeland (1979) has suggested that the dense colonies that *P.humuli* form are responsible for the low rates of parasitism because "Hymenopteran parasitoids generally are best suited to seek out and attack individual aphids, whereas predators, such as Coccinellidae and Syrphidae, are better adapted to attack aphid colonies" (p.150). Van der Ent et al. (2017) suggest that aphid parasitoids will avoid large aphid colonies in order to avoid the large amounts of the sticky honeydew produced. However, in B.C., the cabbage aphid, *Brevicoryne brassicae*, is also found in dense colonies with honeydew, and parasitism frequently occurs (personal communication, Personal communication with Renee Prasad, University of the Fraser Valley).

Figure 24: Parasitoid collected with *Phorodon humuli* from *Prunus* spp.

Entomopathegenic fungus

Of the *P. humuli* killed by beneficial fungus in this 2019 Fraser Valley Field Survey, all were unwinged except for one. This is likely a result of the timing of the fungus and the lifecycle of the aphids. Winged aphids were not seen in fields after July 5, 2019. Species of *Cladosporium* have been studied for their entomopathegenic properties on whiteflies and aphids (Abdel-Baky & Abdel-Salam, 2003). Aphid mortality numbers observed in this 2019 Fraser Valley field survey ranged from 0.02%–25%. The average range in mortality is lower compared to Abdel-Baky & Abdel-Salam (2003), who observed 16.4 to 45.27% mortality by *Cladosporium* spp. While Abdel-Baky & Abdel-Salam (2003) observed higher mortality, the aphids species that were studied did not include *Phorodon humuli*, but rather *Aphis gossypii*, *Aphis craccivora* and *Aphis durantae* (Abdel-Baky & Abdel-Salam, 2003). Future proof of pathogenecy studies would need to be conducted in order to conclude *Cladosporium spp.* was responsible for mortality of *P.humuli* in Fraser Valley hop fields, and also to determine the species of *Cladosporium.*

Contributions to Aphid Control by Natural Enemies

In Woods et al. (2014), populations of coccinellids and aphids were synchronized, but were not able to keep aphid levels below threshold levels. In this study, all aphid predators were low when compared to aphid numbers throughout the season, even in fields with high aphid levels (Figs. 17 & 18). The methods used in this study may have underestimated the species composition and levels. As explained by both Campbell (1978) and Barber et al. (2003), leaf sampling underestimates mobile species because they can either drop off, hide or fall off when leaves are handled (Barber et al., 2003). Pesticides were used in five out of six fields to manage *P. humuli* (Figs. 11 & 12) in this 2019 Fraser Valley survey. Achieving control of *P. humuli* with naturally occurring enemies is not common in commercial hop fields and pesticide intervention is typically required (Copland, 1979; Turner et al., 2011; Weihrauch et al., 2012; Woods et al., 2014).

Multiple factors have been blamed for the lack of success of biological control in managing *P. humuli*, many of which result from the growth habit of hops (Campbell, 1978; Woods et al., 2014). While the hop rhizomes are perennial, the growth above ground is annual. This means that there is only above-ground growth suitable for both aphid and beneficial establishment for a short period each year (Campbell, 1978). The above-ground die back each year does not allow for any overwintering sites for beneficials (Woods et al., 2014). As a result, the natural enemies must re-establish each year, and typically lag behind the arrival of winged aphids in the spring, allowing aphid populations to take off quickly (Campbell, 1978).

The lack of other available aphids for aphid-predators to feed on prior to the arrival of winged aphids into the field has also been attributed to the monoculture environment of hopyards, specifically, the lack of weeds as alternative aphid hosts (Campbell, 1978; Campbell, 2018). Campbell & Cone (1994) note the absence of other aphid species in hopyards prior to the spring arrival of *P. humuli* means there are no food sources available for generalist predators to develop prior to the migration of *P. humuli* into the field. In Fraser Valley fields, *A. fabae* and *M. euphorbiae* were found in hop yards two weeks prior to the arrival of *P. humuli.* While weeds

may have contributed to the presence of other aphid species in Fraser Valley fields, predator development did not catch up before weeds were controlled.

Many beneficial insects benefit from nectar and pollen sources (Van der Ent et al., 2017). Many species of syrphid adults require pollen and nectar for egg production (Van der Ent et al., 2017). Similarily, the fecundity of lacewing adults is linked with both nectar and pollen quality (Van der Ent et al., 2017). Nectar or honeydew can increase both the life cycle and fecundity of parasitoids, and is essential for energy for the parasitoid to carry out its normal functions (Lewis, Stapel, Cortesero, & Takasu, 1998). The term "parasitoid nectar provision hypothesis" was coined by Heimpel & Jervis (2005) and suggests that efficacy of parasitoids can be improved through the provision of nectar sources.

The monocropping of hopyards not only means that there is no plant source to sustain other aphids or pests in order to build predator populations, but it also means that there is also no significant nectar or pollen sources. Grasswitz & James (2009) wrote "without deliberate intervention, non-crop floral resources are often scare in agriculture" (p. 2011). In the case of hopyards, female hops do not produce pollen or nectar, so the crop itself is not contributing any floral resources for predators. This, combined with the lack of permanent weeds, creates an imbalanced system that benefits aphid growth and hinders growth of beneficials.

Recent conservation biological control studies in the eastern USA (Calderwood, Cubins, Vesty, & Darby, 2017), England (Campbell, 2018) and Washington (Grasswitz & James, 2009) have examined the possibility of increasing natural enemies in hopyards through interrow plantings with annual plants. In a companion planting study in England, planting mixes influenced the aphid parasitoids found in hops (Campbell, 2018). More aphid mummies were found in hops under sown with *Brassica juncea* (brown mustard) compared to other treatments (Campbell, 2018).

Experimental Results: Host Transfer Experiments

The third objective was to determine if *P. humuli* had secondary summer hosts other than hops.

Experiment 1: Winged Transfer Study

There were challenges in transferring winged aphids to all hosts, including hops (Appendix F). This was reflected by the different number of aphids surviving transfer at the start of this trial. Host finding for migratory aphids, such as *P. humuli,* involves several factors and can be quite complex. These complexities may have played a role in the challenges experienced in this study when attempting to get *P. humuli* to establish on hosts. Weihrauch & Moreth (2005) explained that the ability of an aphid to identify an appropriate host involves "visual, olfactory, mechanical and gustatory stimuli, with any combination of these being possible" (p. 702). Work has been done to manipulate the hop landscape to interfere with the ability of *P. humuli* to find and colonize hops by both visual and olfactory cues (Campbell & Ridout, 2001; Lösel et al., 1996; Pope, Campbell, Hardie, Pickett, & Wadhams, 2006). A factor that may have interfered with *P. humuli's* ability to colonize the plants was the type of cages used (Fig. 5). The Educational

Science Giant Square Pop-Up Butterfly Cage had two mesh sides and two clear, reflective sides. Reflective row covers are used to deter aphids from entering or landing in certain agricultural fields (Johnson, Bing, & Smith, 1967; Stapleton & Summers, 2002). The reflective siding on these cages may have had a similar effect to that of reflective mulch.

While alates eventually established on hops and populations grew, and declined on nettle and cannabis ('Afghan Kush') (Fig. 19), it is unclear if this was due to suitability of the host or as a result of a combinations of factors which may have impacted or interfered with the aphids' host searching ability.

Experiment 2: Unwinged Transfer Study

While it was difficult to discern clear results in the Winged Transfer Study, the results of Unwinged Transfer Study contributes to the argument that *P. humuli* uses only hops as a summer host (Campbell, 1985; Campbell & Ridout, 2001). The results of this study are in alignment with observations in other hop growing states where cannabis has been legalized. In Oregon, *P. humuli* has not yet been observed on cannabis (Anonymous, 2017). Similarly, there have been no reports of *P. humuli* on cannabis or hemp in Colorado (Anonymous, 2018). It does not appear that cannabis is a suitable summer host of *P. humuli.*

Phorodon humuli and *P. cannabis* are similar in appearance (Cranshaw et al., 2018; Anonymous, 2017), and this has been noted as a reason behind some of the confusion surrounding their respective crop range (Anonymous, 2018). Additionally, cannabis research and pest surveying has not been common because it was not a crop that was grown legally in most locations until recently (Hammon et al., n.d.). The current state of available cannabis pest management information has been described as "incomplete, unreliable, and or simply incorrect" because it has not been written by trained entomologist or pathologists (Hammon et al., n.d., p.1). The shared similarities physical appearance between the two aphids is likely one of the main reasons surrounding the confusion about host ranges of these two pests.

CONCLUSION

Understanding the aphid species composition in hop fields is an important step in approaching aphid control from an integrated pest management perspective. While other aphid species were found in Fraser Valley hop fields, this study confirmed that *P. humuli* is the most important species and it stays in fields past harvest.

Findings to date in the Fraser Valley are consistent with Cranshaw et al., (2018) in that *P. humuli* and *P. cannabis* do not overlap in terms of hosts. *P. cannabis* was not found in any of the six hop fields surveyed in the Fraser Valley in 2019 and *P. humuli* did not establish on cannabis in either two transfer studies when both alates and apetera were used.

This work provides a snapshot of the natural enemies in Fraser Valley hop fields. Ladybird beetles are important predators on leaves while *Orius* spp., was the predominant predator in the

burrs and cones. It is unclear how much these predators are contributing to control of aphids, however, the new knowledge of who the primary predators are in hop fields in this region provides a foundation for future work.

RECOMMENDATIONS

The post-harvest results of this 2019 field survey have practical applications in the management of *P. humuli*. In particular, the new knowledge that *P. humuli* stays, reproduces and continues to feed in Fraser Valley hop fields long after harvest is an opportunity for cultural control. Hop growers should clean up any remaining vegetation in the hop fields after harvest, so that the number of aphids that make it back to *Prunus* spp. to mate can be reduced. This could reduce the amount of aphid pressure in hop fields the following spring.

Research that aims to help hop growers adopt a multi-pronged approach to aphid control should continue. Specifically, based off the findings in this study, future projects should focus on:

1) Strategies for building predator levels in hop fields, especially predators that search in the hop cones. As hop aphids are of particular concern if they enter into the hop cone, predators that can actively seek out aphids in this location are of important consideration for future work. This is also important in the context of the Fraser Valley where two of the four products registered for control of aphids in hop have pre-harvest intervals of 21 days and 31 days (Sivanto Prime Insecticide, n.d.; Beleaf Insecticide, n.d.). This leaves a period where the crop is a vulnerable to aphid infestations. *Orius* spp. were the primary predator found in the burrs and cones in this 2019 field survey. Future studies should focus on this period where there are not many options for protecting the crop.

2) Identifying the diversity of ladybird beetle species in Fraser Valley hop fields. Natural enemies in this 2019 study were not identified to the genus or species level. Given that ladybird beetles were the most abundant predator in Fraser Valley fields on leaves, future work should examine what species are present. Gaining a better understanding of what ladybird species are present is helpful in improving the success of biological controls of aphids in hop fields.

3) Confirming the entomopathegenic fungus present in hop fields through proof of pathogenecy studies. This is needed to conclude *Cladosporium* spp. was responsible for mortality of *P. humuli* in Fraser Valley hop fields in 2019, and also to determine the species of *Cladosporium.*

The transfer experiments conducted in 2019 were exploratory, and there are many opportunities to improve upon the methods for future studies. Future studies should ensure that appropriate cages are used in order to avoid the possible effects of the reflective side material used on the cages in the 2019 transfer experiments. Future transfer experiments should examine not just reproductive abilities of the aphids, but also feeding ability and probing behaviour of the aphids while on different hosts. While these studies provide insight into the host range of *P. humuli*, they do reflect the real-world scenarios aphids face in selecting hosts. For example, in Experiment 2: Unwinged, aptera were put onto cannabis plants. For this to occur in a field situation, hops would need to be undersown with cannabis in order for the aphids to be able to

walk from hops to cannabis. Given that the outdoor cannabis industry and hemp is still developing in the Fraser Valley, future efforts might be best directed to field survey work as these best reflect the realities faced by both hop and cannabis growers.

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WORKS CITED

Abdel-Baky, N. F., & Abdel-Salam, A. H. (2003). Natural incidence of *Cladosporium* spp. As a bio-control agent against whiteflies and aphids in Egypt. *Journal of Applied Entomology*, *127*(4), 228–235. https://doi.org/10.1046/j.1439-0418.2003.00662.x

Agricultural Land Commission. (2014). Panel Regions and Regional Districts. Retrieved from https://www.alc.gov.bc.ca/assets/alc/assets/about-the-alc/alr-and-maps/maps-and-gis/alc_panels_regional_districts.jpg on Feb. 24, 2020

Anonymous. (2017). Pest Alert: Cannabis or Bhang Aphid *Phorodon cannabis*. Oregon Department of Agriculture Fact Sheet. Retrieved from http://www.oregon.gov/ODA/shared/Documents/Publications/IPPM/CannabisAphidAlert.pdf on March 6, 2019

Anonymous. (2018). Cannabis Aphid. Colorado State Hemp Insect Fact Sheet. Retrieved from https://webdoc.agsci.colostate.edu/hempinsects/PDFs/Cannabis%20aphid%20October%202018%20revision%20(1).pdf on March 6, 2019

Aveling, C. (1981). The role of *Anthocoris* species (Hemiptera: Anthocoridae) in the integrated control of the damson-hop aphid (*Phorodon humuli*). *Annals of Applied Biology*, *97* (2), 143–153. https://doi.org/10.1111/j.1744-7348.1981.tb03007.x

Barber, A., Campbell, C.A.M., Crane, H., Darby, P., & Lilley, R. (2003). Cost-benefits of reduced aphicide usage on dwarf hops susceptible and partially resistant to damson-hop aphid. *Annals of Applied Biology*, *143*(1), 35–44. https://doi.org/10.1111/j.1744-7348.2003.tb00266.x

Beleaf Insecticide. (n.d). Pest Management Regulatory Agency Registered Products Database. Pesticide label retrieved from online label search https://pr-rp.hc-sc.gc.ca/1_1/view_label?p_ukid=151838899 on January 19, 2020

Blackman, R.L. & Eastop, V.F. (1984) Aphids on the World's Crops: Identification and Information Guide. John Wiley Sons (p.108-110; 154-155)

Calderwood, L. B., Lewins, S. A., & Darby, H. M. (2015). Survey of Northeastern Hop Arthropod Pests and Their Natural Enemies. *Journal of Integrated Pest Management*, *6*(1). https://doi.org/10.1093/jipm/pmv017

Calderwood, L., Cubins, J., Vesty, D., & Darby, H. (2017). Effect of Drive Row Ground Covers on Hop (Rosales: Cannabaceae) Yard Arthropod Pests in Vermont, USA. *Environmental Entomology*, *46*(2), 183–190. https://doi.org/10.1093/ee/nvw069

Campbell, C. A. M. (1978). Regulation of the damson-hop aphid (*Phorodon humuli* (Schrank)) on Hops (*Humulus lupulus* L.) by Predators. *Journal of Horticultural Science*, *53*(3), 235–242. https://doi.org/10.1080/00221589.1978.11514824

Campbell, C.A.M. (1977). Distribution of damson-hop aphid (*Phorodon humuli*) migrants on hops in relation to hop variety and wind shelter. *Annals of Applied Biology*, *87*(3), 315–325. https://doi.org/10.1111/j.1744-7348.1977.tb01896.x

Campbell, C.A.M. (1983). Antibiosis in Hop (*Humulus lupulus*) to the Damson- Hop Aphid, *Phorodon humuli*. *Entomologia Experimentalis et Applicata*, *33*(1), 57–62. https://doi.org/10.1111/j.1570-7458.1983.tb03233.x

Campbell, C.A.M. (1985). Has the damson-hop aphid an alate alienicolous morph? *Agriculture, Ecosystems & Environment*, *12*(3), 171–180. https://doi.org/10.1016/0167-8809(85)90108-2

Campbell, C.A.M., & Cone, W. W. (1994). Influence of Predators on Population Development of *Phorodon humuli* (Homoptera: Aphididae) on Hops. *Environmental Entomology*, *23*(6), 1391–1396. https://doi.org/10.1093/ee/23.6.1391

Campbell, C.A.M., & Ridout, M. S. (2001). Effects of plant spacing and interplanting with oilseed rape on colonisation of dwarf hops by the damson-hop aphid, *Phorodon humuli*. *Entomologia Experimentalis et Applicata*, *99*(2), 211–216. https://doi.org/10.1046/j.1570-7458.2001.00819.x

Campbell, C.A.M. (2018). Influence of companion planting on damson hop aphid *Phorodon humuli*, two spotted spider mite *Tetranychus urticae*, and their antagonists in low trellis hops. *Crop Protection*, *114*, 23–31. https://doi.org/10.1016/j.cropro.2018.08.014

Copland, M. J. W. (1979). Hymenoptera in hop gardens, with particular reference to parasitoids associated with the damson-hop aphid *Phorodon Humuli*. *Entomologia Experimentalis et Applicata*, *25*(2), 146–152. https://doi.org/10.1111/j.1570-7458.1979.tb02864.x

Cranham, J. E. (1982). Integrated control of damson-hop aphid, *Phorodon humuli*, on English hops: A review of recent work. *Agriculture and Environment*, *7*(1), 63–71. https://doi.org/10.1016/0304-1131(87)90007-5

Cranshaw, W. S., Halbert, S.E., Favret, C., Britt, K., & Miller, G. (2018). *Phorodon cannabis* Passerini (Hemiptera: Aphididae), a newly recognized pest in North America found on industrial hemp. *Insecta Mundi, 0662*, 1-12. DOI: urn:lsid:zoobank.org:pub:21F39645-D23B-4532-AA49-E07C46B2ECF3

Denman, R. (2008). Brewer's Gold Online Exhibit. Chilliwack Museum and Archives. Retrieved from http://www.chilliwackmuseum.ca/hops/BG_Main.html on Dec. 7, 2019

Dorschner, K. W., & Baird, C. R. (1988). Susceptibility of hop to *Phorodon humuli*. *Entomologia Experimentalis et Applicata*, *49*(3), 245–250. https://doi.org/10.1111/j.1570-7458.1988.tb01185.x

Dorschner, K. W., Feng, M.G., & Baird, C. R. (1991). Virulence of an Aphid-Derived Isolate of *Beauveria bassiana* (Fungi: Hyphomycetes) to the Hop Aphid, *Phorodon humuli* (Homoptera: Aphididae). *Environmental Entomology*, *20*(2), 690–693. https://doi.org/10.1093/ee/20.2.690

Dreves, A. J., & Walsh, D. B. (2015). Hop Aphid**.** In D. B. Walsh, D.H. Gent, J.D. Barbour, R.A. Boydston, A.E. George, D.G. James & J.R. Sirrine (Eds), *Field Guide for Integrated Pest Management in Hops* (p. 47-48). Retrieved from https://www.usahops.org/resources/field-guide.html on Dec. 8, 2019

Eastwell, K. C., & Barbara, D. Z. (2015). Virus and Viroid Diseases**:** Carlavirus Complex: *American hop latent virus*, *Hop latent virus*, and *Hop mosaic virus***.** In D. B. Walsh, D.H. Gent, J.D. Barbour, R.A. Boydston, A.E. George, D.G. James and J.R. Sirrine (Eds), *Field Guide for Integrated Pest Management in Hops* (p.38-41). Retrieved from https://www.usahops.org/resources/field-guide.html on Dec. 8, 2019

Evans, G. (2004). Brewers Gold – Hops History in BC. In R. Keen, *Small-Scale and Organic Hops Production* (pp. 4-5). Retrieved from https://cdn.shopify.com/s/files/1/1126/3316/files/HopsManual.pdf?17901602041566931036 on Dec. 7, 2019

Fraser Valley Regional District (2017). Regional Snapshot Series: Agriculture Agricultural Economy in the Fraser Valley Regional District. Retrieved from https://www.fvrd.ca/assets/Government/Documents/AgricultureSnapshot.pdf on Feb. 15, 2020

Filotas, M. (2012). *Hop Pest Management: What's bugging your bines?* Ontario Ministry of Agriculture, Food and Rural Affairs (OMAFRA). Retrieved from https://onspecialtycrops.files.wordpress.com/2013/04/hops-scouting-presentation-2012.pdf on June 16, 2019

Gillespie, D. R., & Quiring, D. M. J. (2006). *Orius insidiosus* (Hemiptera: Anthocoridae) in the lower Fraser Valley, British Columbia, Canada,. *The Canadian Entomologist*, *138*(4), 590–593. https://doi.org/10.4039/n06-802

Grasswitz, T. R., & James, D. G. (2009). Influence of hop yard ground flora on invertebrate pests of hops and their natural enemies. *Journal of Applied Entomology*, *133*(3), 210–221. https://doi.org/10.1111/j.1439-0418.2008.01336.x

Hammon, B., Rizza, J., & Dean, D. (n.d.). *Current Impacts of Outdoor Growth of Cannabis in Colorado.* Colorado State University Extension Fact Sheet N. 0.308. Retrieved from https://mountainscholar.org/bitstream/handle/10217/185141/AEXT_003082015.pdf?sequence=1 on Feb. 14, 2020

Heimpel, G. E., & Jervis, M. A. (2005). Does floral nectar improve biological control by parasitoids? In F. L. Wackers, P. C. J. van Rijn & J. Bruin (Eds), *Plant-Provided Food for Carnivorous Insects,* Cambridge University Press (p.267- 293). Retrieved from https://books.google.ca/books?hl=en&lr=&id=7j1WyqPAARwC&oi=fnd&pg=RA4-PA267&dq=parasitoid+nectar+provision+hypothesis&ots=Flz8UfLngi&sig=5uhTTyiwA285dlgfGuiITIZbwwk#v=onepage&q=parasitoid%20nectar%20provision%20hypothesis&f=false on Feb. 27, 2020

Herring, J. L. (1966). The Genus *Orius* of the Western Hemisphere (Hemiptera: Anthocoridae). *Annals of the Entomological Society of America*, *59*(6), 1093–1109. https://doi.org/10.1093/aesa/59.6.1093

James, D. G. & Dreves, A. J. (2015). Beneficial Arthropods and Pathogens**.** In D. B. Walsh, D. H. Gent, J. D. Barbour, R.A. Boydston, A.E. George, D. G. James & J. R. Sirrine (Eds), *Field Guide for Integrated Pest Management in Hops* (p.71-86). Retrieved from https://www.usahops.org/resources/field-guide.html on Dec. 8, 2019

Johnson, G. V., Bing, A., & Smith, F. F. (1967). Reflective Surfaces Used to Repel Dispersing Aphids and Reduce Spread of Aphid-Borne Cucumber Mosaic Virus in Gladiolus Plantings. *Journal of Economic Entomology*, *60*(1), 16–18. https://doi.org/10.1093/jee/60.1.16

Lago, P. K., & Stanford, D. F. (1989). Phytophagous insects associated with cultivated marijuana, *Cannabis sativa*, in Northern Mississippi. *Journal of Entomological Science*, *24*(4), 437–445. https://doi.org/10.18474/0749-8004-24.4.437

Lerin, J., & Badenhausser, I. (1995). Influence of the leaf curling plum aphid (*Brachycaudus helichrysi*) on stem diameter, seed yield, and their relationship, in sunflower. *The Journal of Agricultural Science*, *125*(2), 211–221. https://doi.org/10.1017/S0021859600084331

Lewis, W. J., Stapel, J. O., Cortesero, A. M., & Takasu, K. (1998). Understanding How Parasitoids Balance Food and Host Needs: Importance to Biological Control. *Biological Control*, *11*(2), 175–183. https://doi.org/10.1006/bcon.1997.0588

Lizotte, E., Hodgson, E., & Filotas, M. (2017). Michigan State University Extension. Hop Scouting Pocket Guide for the U.S. Upper Midwest and Northwest, and Eastern Canada.

Lorenzana, A., Hermoso de Mendoza, A., Seco, M. V., & Casquero, P. A. (2010). Population development of *Phorodon humuli* and predators (*Orius* spp.) within hop cones: Influence of aphid density on hop quality. *Crop Protection*, *29*(8), 832–837. https://doi.org/10.1016/j.cropro.2010.04.014

Lösel, P. M., Lindemann, M., Scherkenbeck, J., Campbell, C. A. M., Hardie, J., Pickett, J. A., & Wadhams, L. J. (1996). Effect of primary-host kairomones on the attractiveness of the hop-aphid sex pheromone to Phorodon humuli males and gynoparae. *Entomologia Experimentalis et Applicata*, *80*(1), 79–82. https://doi.org/10.1111/j.1570-7458.1996.tb00890.x

Loxdale, H. D., Brookes, C. P., Wynne, I. R., & Clark, S. J. (1998). Genetic variability within and between English populations of the damson–hop aphid, *Phorodon humuli* (Hemiptera: Aphididae), with special reference to esterases associated with insecticide resistance. *Bulletin of Entomological Research*, *88*(5), 513–526. https://doi.org/10.1017/S0007485300026031

McPartland, J., Clarke, R., & Watson, D. (2000). Hemp Diseases and Pests Management and Biocontrol. CAB International (p.31-35)

Morrison, H. E. (1958). *Hop pest control* (Oregon State University). Agricultural Experiment Station, Oregon State College. Circular of Information 589 retrieved from https://ir.library.oregonstate.edu/concern/administrative_report_or_publications/rb68xc15q on Feb. 14, 2019

Patterson, R., & Ramirez, R. (2016). *Aphid Natural Enemies and Biological Control*. Utah State University Extension. Retrieved from https://digitalcommons.usu.edu/cgi/viewcontent.cgi?article=1813&context=extension_curall on Feb. 25, 2020

Pérez, N., Seco, M. V., Valenciano, J. B., & Lorenzana, A. (2007). Use of suction-trap and Moericke traps for monitoring the migration of damson-hop aphid (*Phorodon humuli*) (Hemiptera, Aphididae). *New Zealand Journal of Crop and Horticultural Science*, *35*(4), 455–461. https://doi.org/10.1080/01140670709510214

Pest Management Regulatory Agency Online Label Search. (n.d.). Retrieved from https://pr-rp.hc-sc.gc.ca/ls-re/index-eng.php on January 19, 2020

Pike, K. S., & Starý, P. (1995). Binodoxys conei sp. N. (Hymenoptera, Braconidae, Aphidiinae), a New Parasitoid of Hop Aphid, Phorodon humuli (Schrank) in Washington State. *Journal of the Kansas Entomological Society*, *68*(4), 444–450. JSTOR

Pike, K. S., & Starý, P., Miller, T., Graf, D., Allison, D., Boydston, L & Miller, R. (2000). Aphid Parasitoids (Hymenoptera, Braconidae, Aphidiinae) of Northwest USA. *Proceedings of the Entomological Society of Washington*, *102*(3), 688-740. https://www.biodiversitylibrary.org/item/54765#page/1/mode/1up

Pope, T. W., Campbell, C. A. M., Hardie, J., Pickett, J. A., & Wadhams, L. J. (2006). Interactions Between Host-plant Volatiles and the Sex Pheromones of the Bird Cherry-oat Aphid, Rhopalosiphum padi and the Damson-hop Aphid, Phorodon humuli. *Journal of Chemical Ecology*, *33*(1), 157–165. https://doi.org/10.1007/s10886-006-9199-4

Prasad, R. P., Kabaluk, J. T., Meberg, H. P., Bevon, D. A., & Henderson, D. E. (2009). Seasonal and Spatial Occurrence of Aphid Natural Enemies in Organic *Brassica* Fields: Diversity, Phenology, and Reproduction. *Journal of Sustainable Agriculture*, *33*(3), 336–348. https://doi.org/10.1080/10440040902773178

Province of British Columbia. (n.d). Factsheet: Growing non-medical cannabis at home in British Columbia. Retrieved from https://www2.gov.bc.ca/assets/gov/public-safety-and-emergency-services/public-safety/cannabis/grow-nonmedical-cannabis-factsheet.pdf on Feb. 29, 2020.

Smith, K. (1951). A textbook of Agricultural Entomology. Cambridge University Press (p.66). Retrieved from https://www.cabdirect.org/cabdirect/abstract/19311000113 on March 6, 2019

Stapleton, J. J., & Summers, C. G. (2002). Reflective mulches for management of aphids and aphid-borne virus diseases in late-season cantaloupe (*Cucumis melo* L. var. Cantalupensis). *Crop Protection*, *21*(10), 891–898. https://doi.org/10.1016/S0261-2194(02)00055-8

Sirrine, J. R. (2009). Sustainable Hop Production in the Great Lakes Region. *Michigan State Univesrity Extension*, 8

Sivanto Prime Insecticide. (n.d.) Pest Management Regulatory Agency Registered Products Database. Pesticide label retrieved from online label search https://pr-rp.hc-sc.gc.ca/1_1/view_label?p_ukid=151840237 on January 19, 2020

Taimr, L., & Kŕíž, J. (1978). Stratiform drift of the hop aphid (*Phorodon humuli* Schrank). *Zeitschrift für angewandte Entomologie*, *86*(1–4), 71–79. https://doi.org/10.1111/j.1439-0418.1978.tb01913.x

Taylor, L. R., Woiwod, I. P., & Taylor, R. A. J. (1979). The Migratory Ambit of the Hop Aphid and its Significance in Aphid Population Dynamics. *The Journal of Animal Ecology*, *48*(3), 955. https://doi.org/10.2307/4207

Turner, S. F., Benedict, C. A., Darby, H., Hoagland, L. A., Simonson, P., Sirrine, J. R., & Murphy, K. M. (2011). Challenges and Opportunities for Organic Hop Production in the United States. *Agronomy Journal*, *103*(6), 1645. https://doi.org/10.2134/agronj2011.0131

USDA National Agricultural Statistics Service. (2019). National Hop Report. Retrieved from https://www.usahops.org/img/blog_pdf/267.pdf on Feb. 29, 2020

Van der Ent, S., Knapp, M., Klapwijk, J., Moerman, E., van Schelt, J., & de Weert, S. (2017). Knowing and Recognizing: The Biology of Pests, Diseases and their Natural Solutions. Koppert B.V. (p.174-199).

Walsh, D. B. (2015). Pesticide Toxicology and Selectivity**.** In D. B. Walsh, D. H. Gent, J. D. Barbour, R.A. Boydston, A. E. George, D. G. James & J. R. Sirrine (Eds), *Field Guide for Integrated Pest Management in Hops* (p.6-7). Retrieved from https://www.usahops.org/resources/field-guide.html on Dec. 8, 2019

Weihrauch, F. (2005). Evaluation of a damage threshold for two-spotted spider mites, *Tetranychus urticae* Koch (Acari: Tetranychidae), in hop culture. *Annals of Applied Biology*, *146*(4), 501–509. https://doi.org/10.1111/j.1744-7348.2005.040163.x

Weihrauch, F, Baumgartner, A., Felsl, M., Kammhuber, K., & Lutz, A. (2012). The Influence of Aphid Infestation during the Hop Growing Season on the Quality of Harvested Cones. *Brewing Science*, *65*, 9. https://www.researchgate.net/profile/Florian_Weihrauch/publication/259356935_The_Influence_of_Aphid_Infestation_during_the_Hop_Growing_Season_on_the_Quality_of_Harvested_Cones/links/0046352b2b326d2300000000.pdf

Weihrauch, Florian, & Moreth, L. (2005). Behavior and Population Development of *Phorodon humuli* (Schrank) (Homoptera: Aphididae) on Two Hop Cultivars of Different Susceptibility. *Journal of Insect Behavior*, *18*(5), 693–705. https://doi.org/10.1007/s10905-005-7020-9

White, G. B. (1948). The Development of the Eastern Fraser Valley. In W.E. Ireland (Ed.), *The British Columbia Historical Quarterly (October)* (p. 259-291). Retrieved from http://www.library.ubc.ca/archives/pdfs/bchf/bchq_1948_4.pdf on Dec. 7, 2019

Woods, J. L., Dreves, A. J., James, D. G., Lee, J. C., Walsh, D. B., & Gent, D. H. (2014). Development of Biological Control of *Tetranychus urticae* (Acari: Tetranychidae) and *Phorodon humuli* (Hemiptera: Aphididae) in Oregon Hop Yards. *Journal of Economic Entomology*, *107*(2), 570–581. https://doi.org/10.1603/EC13488

Worner, S. P., Tatchell, G. M., & Woiwod, I. P. (1995). Predicting spring migration of the damson-hop aphid *Phorodon humuli* (Homoptera: Aphididae) from historical records of host-plant flowering phenology and weather. *The Journal of Applied Ecology*, *32*(1), 17. https://doi.org/10.2307/2404412

Wright, L. C., Cone, W. W., & James, D. G. (2005). Sources of Spring and Fall Hop Aphid, *Phorodon humuli* (Schrank), (Homoptera: Aphididae) Migrants in South Central Washington. *Journal of the Entomological Society of British Columbia*, *102*(0), 21–26.

Wright, L. C., & James, D. G. (2001). Parasitoids of the Hop Aphid (Homoptera: Aphididae) on Prunus during the Spring in Washington State. *Journal of Agricultural and Urban Entomology*, *18*(3), 7

Wright, L. C., Pike, K. S., Allison, D., & Cone, W. W. (1995). Seasonal Occurrence ofAlate Hop Aphids (Homoptera: Aphididae) in Washington State. *Journal of Agricultural Entomoloy*, *12*(1), 12

APPENDICES

Appendix A: Aphid identification reports for Field Survey NIS/SIN lot: 2019-187-190 (Agriculture and Agri-Food Canada)

Agriculture and Agri-Food Canada Agriculture et Agroalimentaire Canada

NIS/SIN lot : **2019-187**

your file/votre dossier :
BCMA-Tracy/Emily

Systematic Entomology
IDENTIFICATION RECORD

Entomologie systématique
FICHE D'IDENTIFICATION

your number votre numéro	our number notre numéro	**Identification and remarks Identification et remarques**	No. of specimens Nbre de specimens	Identification incomplete Identification incomplète	Retained Retenus
		HEMIPTERA: APHIDIDAE			
Sept		*Phorodon humuli* (Schrank)	45		yes
Oct		*Ph. humuli*	12		yes
Raymond June 7		*Ph. humuli*	2		yes

Identified by: / Identifié par: **Eric Maw** Eric.Maw@agr.gc.ca Date: **2019.11.01**

Agriculture and Agri-Food Canada | Agriculture et Agroalimentaire Canada

NIS/SIN lot : **2019-188**

your file/votre dossier :
BCMA-Tracy/Emily

Systematic Entomology
IDENTIFICATION RECORD

Entomologie systématique
FICHE D'IDENTIFICATION

your number votre numéro	our number notre numéro	**Identification and remarks Identification et remarques**	No. of specimens Nbre de specimens	Identification incomplete Identification incomplète	Retained Retenus
		HEMIPTERA: APHIDIDAE			
McMath may 13		*Aphis fabae* Scopoli	2		yes
McMath june 21		*A. fabae*	26		yes
Cole Rd june 21		*A. fabae*	12		yes

Identified by: / Identifié par: **Eric Maw** Eric.Maw@agr.gc.ca Date: **2019.11.01**

Agriculture and Agri-Food Canada | Agriculture et Agroalimentaire Canada

NIS/SIN lot : **2019-189**

your file/votre dossier :
BCMA-Tracy/Emily

Systematic Entomology
IDENTIFICATION RECORD

Entomologie systématique
FICHE D'IDENTIFICATION

your number votre numéro	our number notre numéro	**Identification and remarks Identification et remarques**	No. of specimens Nbre de specimens	Identification incomplete Identification incomplète	Retained Retenus
		HEMIPTERA: APHIDIDAE			
McMath may 13		*Macrosiphum euphorbiae* (Thomas)	1		yes
Royalwood May 10		*M. euphorbiae*	2		yes
Topps May 10		*M. euphorbiae*	1		yes

Identified by: / Identifié par: **Eric Maw** Eric.Maw@agr.gc.ca Date: **2019.11.01**

Agriculture and Agri-Food Canada | Agriculture et Agroalimentaire Canada

NIS/SIN lot : **2019-190**

your file/votre dossier :
BCMA-Tracy/Emily

Systematic Entomology
IDENTIFICATION RECORD

Entomologie systématique
FICHE D'IDENTIFICATION

your number votre numéro	our number notre numéro	**Identification and remarks Identification et remarques**	No. of specimens Nbre de specimens	Identification incomplete Identification incomplète	Retained Retenus
		HEMIPTERA: APHIDIDAE			
Royalwood june 21		*Brachycaudus helichrysi* (Kaltenbach)	2		yes
Royalwood May 10		*Macrosiphum euphorbiae* (Thomas)	2		yes
Cole Rd May 10		*Aphis fabae* Scopoli	1		yes
Topps June 7		no aphid; debris only found			

Identified by: / Identifié par: **Eric Maw** Eric.Maw@agr.gc.ca Date: **2019.11.01**

Appendix B: Map of assigned plants to cages by R for Experiment 1: Winged Transfer

N ↑

H3	H4
C4	N3
H1	N2
N4	C3
C1	H2
C2	N1

H= Hops – ‘Cascade’
N= Nettle
C= Cannabis – ‘Afghan Kush’

Appendix C: Aphid identification report from *Prunus* spp. for rearing - Agriculture and Agri-Foods Canada

NIS/SIN lot : **2019-070**
your file/votre dossier :

Systematic Entomology
IDENTIFICATION RECORD

Entomologie systématique
FICHE D'IDENTIFICATION

your number votre numéro	our number notre numéro	**Identification and remarks** **Identification et remarques**	No. of specimens Nbre de specimens	Identification incomplete Identification incomplète	Retained Retenus
		HEMIPTERA: APHIDIDAE			
April 12		*Hyalopterus pruni* (fundatrix)	1		Yes
April 12		*Phorodon humuli* (fundatrix & nymphs)	14		Yes
April 19		*Phorodon humuli* (fundatrices, apterous viviparae & nymphs)	27		Yes

Identified by:
Identifié par: **Bryan Brunet** bryan.brunet@canada.ca Date: **2018.05.10**

This information is being collected by Agriculture and Agri-Food Canada for the purpose of Research. Information may be accessible or protected as required under the provisions of the Access to Information Act.

Les renseignements sont recueillis par Agriculture et Agroalimentaire Canada pour fins de recherches. Les renseignements peuvent être accessibles ou protégés selon ce que prescript la Loi sur l'accès a l'nformation.

Appendix D: Map of assigned plants to cages by R for Experiment 2: Unwinged Transfer

N ⬆

CAK1	H2
CSSB1	CSSB3
CSSB2	H1
CAK3	H3
CAK2	CAK4
CSSB4	H4

H= Hops – ‘Cascade’
CAK= Cannabis – ‘Afghan Kush’
CSSB = Cannabis – ‘Strawberry Switchblade’

Appendix E: *Cladosporium* spp. identification report - Agriculture and Agri-Foods Canada

Agriculture and Agri-Food Canada | Agriculture et Agroalimentaire Canada

FUNGAL IDENTIFICATION REPORT – FICHE D'IDENTIFICATION

Lot No –No de L'envoi
NFIS5247-5248

CLIENT
NAME-NOM: Todd Kabaluk

ORGANISATION: Agriculture and Agri-Food Canada
ADDRESS – ADDRESSE: PO BOX 1000, 6947 Highway 7, Agassiz, BC V0M 1A0
TEL-TÉL: 604-796-6083
FAX-TELECOPIEUR: 604-796-6133
EMAIL- COURRIER ÉLECTRONIQUE: Todd.Kabaluk@canada.ca

Project-Projet: #

Laboratory File No. – No de fiche du laboratoire:

NOTE: Your material may be deposited in the National Mycological Herbarium (DAOM) or in the Canadian Collection of Fungal Cultures (CCFC)
REMARQUE: Votre matériel peut être déposé à l'Herbier national de mycologie (DAOM) ou à la Collection de cultures fongiques canadiennes (CCFC)

Description of material/ description du materiel provided by the client: Two (2) petri plates containing fungal specimens. Plate#1 (NFIS 5247) was labelled as Blueberry Aphid, subcultured Aug 13/19. Plate#2 (NFIS 5248) was labelled as Hops Aphid, subcultured Aug 13/19. Assigned identifier NFIS numbers 5247-5248.

IDENTIFICATIONS:

2019 NFIS 5247-5248: Two specimens from Todd Kabaluk, Agriculture and Agri-Food Canada, Agassiz, B.C.

The fungi were identified as follows:

NFIS5247 = Plate#1 was identified as *Fusarium oxysporum* species complex. A lot of the Fusarium species are closely related and the newest thinking is that there is a lot of cross mating between species, hence species complex. When the sequences are analyzed with an NCBI BLAST search, the predominant hit appears to be *F. oxysporum f. sp. lycopersici*, based on ITS and 28S sequences. In order to positively identify this sample as *Fusarium oxysporum* microscopically (to confirm the genetic analysis) would require a Fusarium expert. Our resident expert, Keith Seifert, has recently retired. Microscopic identification to species level would require that the fungus be grown in SNA, in order to encourage sporulation, prior to being examined by a *Fusarium* expert. It is safe to say that the identification, based on a genetic analysis, belongs to *Fusarium oxysporum* species complex, and is likely *Fusarium oxysporum f. sp. lycopersici*, based on ITS and 28S sequences. These genes are well documented and preserved in fungi. *Fusarium oxysporum f. sp. Lycopersici* causes Fusarium wilt in tomato plants.

NFIS5248 = Plate#2 was identified as *Cladosporium* sp., based on the well characterized ITS and 28S genes, and compared with result within an NCBI BLAST search. Further, Jonathan Mack examined the plate microscopically and confirmed that the culture was in fact *Cladosporium* sp. Unfortunately, it was not possible to identify the submission to species level based on the genetic analysis, as there were different conclusion that could be drawn based on the results of the 2 genes analyzed. It is possible that the database may have erroneous entries as well, as we can only assume that the species tagged to the sequences that were uploaded are true to the species, and not misidentified. The identification of NFIS5248 could not be confirmed microscopically as no sporulating structures were noted.

Appendix F: Description of failed methods for transferring alates Experiment 1: Winged Transfer

Notes on failed attempts to transfer aphids to hosts (winged trial)

First application: Paintbrush transfer

- May 21: Moved aphids to plants via paintbrush (10 aphids per plant);
- May 22: Counted aphids on plants again, and topped up aphids where they had not survived to ensure 10 per plant;
- May 24: Checked plants. No aphids survived the paintbrush method on any plants.

Second application: Leaf transfer

- May 25: Re-checked plants to ensure no surviving aphids from attempted paintbrush transfer;
- May 25: Re-infested plants via leaves from rearing cages (counted aphids on leaf and recorded numbers, placed leaves into cages overnight);
- May 26: Checked plants. No aphids survived the leaf transfer method on any plants.

Third application: Bulk container transfer

- May 27: Re-checked plants to ensure no surviving aphids from previous attempted transfer methods
- May 27: Collected aphids that were on sides (inside) of rearing cages (aphids had started to aggregate on the inside of the cages). Took a plastic container, opened rearing cage, and tapped on side of cage to dislodge the aphids into the container. Put mesh lid on container, and moved to cage with host plant for aphids. Removed mesh lid and closed cage. Let aphids disperse themselves from plastic container for 2 days.
- May 30: Checked plants. No aphids survived this method of transfer to any plants.

Appendix G: 2019 fungicide records for ‘Cascade’ fields

Farm/Site 1 (‘Cascade’)	Date of application	Active Used	Tradename
No fungicides used in 2019			

Farm/Site 4 (‘Cascade’)	Date of application	Active Used	Tradename
	June 6	Fluopicolide	Presidio Fungicide
	June 6	Boscalid & Pyraclostrobin	Pristine WG Fungicide
	June 17	Dimethomorph	Forum
	June 17	Cyazofamid	Torrent 400 SC Fungicide
	June 28	Mandipropamid	Revus Fungicide
	June 28	Metrafenone	Vivando SC Fungicide
	July 11	Fluopyram & Trifloxystrobin	Luna Sensation
	August 1	Fluopyram & Trifloxystrobin	Luna Sensation
	August 1	Potassium bicarbonate	Milstop Foliar Fungicide
	August 1	Cyazofamid	Torrent 400 SC Fungicide

Farm/Site 6 (‘Cascade’)	Date of application	Active Used	Tradename
	May 1	*Reynoutria sachalinensis*	Regalia
	May 13	*Reynoutria sachalinensis*	Regalia
	May 30	Ametoctradin & Dimethomorph	Zampro
	June 10	Ametoctradin & Dimethomorph	Zampro
	June 20	Ametoctradin & Dimethomorph	Zampro
	July 2	Mandipropamid	Revus Fungicide
	July 15	Potassium bicarbonate	Milstop Foliar Fungicide
	August 5	Potassium bicarbonate	Milstop Foliar Fungicide

Appendix H: 2019 fungicide records for ‘Triple Pearle’ fields

Farm/Site 2: ‘Triple Pearle’	**Date of application**	**Active Used**	**Tradename**
	May 27	Cyazofamid	Torrent 400 SC Fungicide
	June 12	Ametoctradin & Dimethomorph	Zampro
	June 12	Metrafenone	Vivando SC Fungicide
	June 12	Cyazofamid	Torrent 400 SC Fungicide
	July 3	Metrafenone	Vivando SC Fungicide

Farm/Site 3: ‘Triple Pearle’	**Date of application**	**Active Used**	**Tradename**
	May 27	Cyazofamid	Torrent 400 SC Fungicide
	June 6	Ametoctradin & Dimethomorph	Zampro
	June 6	Cyazofamid	Torrent 400 SC Fungicide
	June 10	Metrafenone	Vivando SC Fungicide
	June 10	Ametoctradin & Dimethomorph	Zampro

Farm/Site 5: ‘Triple Pearle’	**Date of application**	**Active Used**	**Tradename**
	May 25	Cyazofamid	Torrent 400 SC Fungicide
	June 13	Ametoctradin & Dimethomorph	Zampro
	June 21	Cyazofamid	Torrent 400 SC Fungicide
	July 4	Metrafenone	Vivando SC Fungicide