Pea aphid dropping behavior diminishes foraging efficiency of a predatory ladybeetle

Devon L. Francke, Jason P. Harmon*, Chad T. Harvey[†] & Anthony R. Ives

Department of Zoology, University of Wisconsin, Madison, WI 53706, USA

Abstract

Antipredator defensive behaviors are a well-studied and often crucial part of prey life histories, but little has been done to quantify how such behaviors affect natural enemies, their foraging, and their effectiveness as biological control agents. We explored how the generalist predatory coccinellid *Harmonia axyridis* Pallas (Coleoptera: Coccinellidae) affects the dropping behavior of the pea aphid, *Acyrthosiphon pisum* (Harris) (Homoptera: Aphididae), and in turn, how that defensive behavior affects the foraging efficiency of the predator. Experimental arenas that allowed or prevented pea aphid dropping were compared to determine how dropping influences the foraging of multiple life stages of *H. axyridis*: second instars, fourth instars, and adults. Dropping reduced predation on aphids by all ladybeetle life stages. Despite older predators inducing more dropping, aphid dropping reduced predation by approximately 40% across all ladybeetle life stages. Aphid dropping and predator consumption of aphids were both correlated with how much the predator moved, which also increased with predator life stage. We suggest that the high rates of dropping induced by *H. axyridis* and the subsequent decrease in *H. axyridis* foraging efficiency may partially explain why *H. axyridis* is less effective at controlling pea aphids than it is at controlling other aphid species that do not drop.

Introduction

Herbivorous insects have evolved many different defensive strategies in response to the risk of mortality from predators and parasitoids (Gross, 1993; Chau & Mackauer, 1997; Braendle & Weisser, 2001). These defensive strategies protect herbivores while potentially reducing natural enemy effectiveness at suppressing pest densities. Induced defenses have been well-studied in predator–prey systems, especially in systems with arthropods (reviewed in Dill et al., 1990; Andrade & Roitberg, 1995; Kunert et al., 2005). These studies have demonstrated the benefit of antipredator behaviors for prey, and in so doing, have proposed a subsequent detrimental effect to the predator. Yet, little has been done to quantify the negative effect of antipredator behaviors on arthropod predator foraging or to understand how such effects may vary among predators.

Pea aphids, *Acyrthosiphon pisum* (Harris) (Homoptera: Aphididae), are a model system for the study of defensive

behaviors. Their large repertoire of defensive strategies includes morphological, social, chemical, and behavioral defenses (Losey & Denno, 1998a; Braendle & Weisser, 2001; Gish & Inbar, 2006; Nelson & Rosenheim, 2006). The behavioral defenses of pea aphids include kicking with its hind legs, walking away, synchronized twitching, and attacking with its frontal horns (Chau & Mackauer, 1997; Losey & Denno, 1998a; Villagra et al., 2002). The most common defensive behavior of pea aphids, however, is dropping from the plant when in immediate danger (Roitberg et al., 1979; Losey & Denno, 1998a,b; and references therein).

Pea aphid dropping is beneficial, because it immediately reduces an aphid's risk of being consumed, but dropping can also be costly and, therefore, may be used selectively. Costs of dropping from a plant may include susceptibility to desiccation, starvation, and predation on the ground (Roitberg & Myers, 1978, 1979; Dill et al., 1990; Gish & Inbar, 2006). A number of factors may influence whether these risks are worth taking. Such factors include the quality of the plant it drops from, the risk of mortality in the new microhabitat it drops to (Losey & Denno, 1998a), and

*Correspondence: E-mail: jharmon@wisc.edu
[†]Present address: Great Lakes Institute for Environmental Research, University of Windsor, Windsor, ON, Canada.

weather conditions (Honěk et al., 1998). Perhaps the most important factor in determining whether to drop is the magnitude of the immediate risk from a predator on the plant (Losey & Denno, 1998a). For example, larger predators, such as coccinellids, induce a large amount of pea aphid dropping, possibly because the aphid associates coccinellids' high consumption rate with a greater risk (Losey & Denno, 1998a,b).

One of the most common pea aphid predators in the mid-western USA is the Asian multi-colored ladybeetle, *Harmonia axyridis* Pallas (Coleoptera: Coccinellidae). This generalist predator consumes pea aphids and can sometimes reach high abundance in alfalfa and nearby crops, thereby giving it the potential to influence pea aphid populations (Snyder & Ives, 2003). This ladybeetle is a generalist predator consuming many species of aphids, other herbivores, other predators, and even conspecifics (Hajek & Dahlsten, 1987; Koch, 2003; Snyder & Ives, 2003).

Harmonia axyridis adults and all instars consume pea aphids (Ueno, 2003), but at different rates and possibly with different foraging behaviors. Different ladybeetle life stages should therefore present different risks to pea aphids and consequently may invoke different aphid behavioral responses. Here, we quantify the dropping behavior induced by different ladybeetle life stages and compare the foraging efficiency of each life stage in experimental arenas where aphids are allowed to drop to those where aphids are not allowed to drop. We further suggest how differences in the foraging behavior of each life stage influence dropping rates.

Materials and methods

To quantify the effects of pea aphid dropping behavior on foraging of three *H. axyridis* life stages, we performed experiments manipulating the ability of aphids to drop from plants. Three-dimensional (3-D) arenas allowed pea aphids to freely drop from the plant, whereas two-dimensional (2-D) arenas allowed predators and prey to interact without prey dropping. We compared the results of experimental replicates in 3-D and 2-D arenas to determine the effects of pea aphid dropping on predator foraging.

Three-dimensional arenas

To quantify the effect of aphid dropping on predation, we subjected aphids in 3-D experimental arenas to one of four treatments: one adult predator, one fourth instar, one second instar, or a no predator control. Three-dimensional arenas were designed to allow pea aphids to drop freely from a host plant while allowing us to accurately count remaining aphids. Each arena included a single, excised fava bean plant [*Vica faba* L. (Fabales: Fabaceae)] (18–25 cm tall). To help improve count accuracy, excised

plants were kept in water-filled vials rather than in soil. Plants were placed in cylindrical mylar sleeves (35 cm in height × 18 cm in diameter) with mesh windows, tops, and bottoms. We used Fluon® (a slippery substance that insects cannot walk on; Northern Products, Inc., Woonsocket, RI, USA) around the base of the mylar sleeve and the vial to prevent dropped aphids from climbing back on the plant. We inspected each arena approximately every 5 min to make sure no predators were feeding on dropped aphids. Larval ladybeetles were never found on the bottom mesh of the arena.

We placed 30 pea aphids on each plant and gave them 10–15 min to acclimate before a predator was introduced to the arena. Half of the aphids were second and third instars, referred to as 'juveniles', and the other half were fourth instar to adult aphids, referred to as 'adults'. We included different sized prey, because previous work suggested that different pea aphid instars may drop at different rates (e.g., Gish & Inbar, 2006). Aphids were from very large laboratory colonies maintained on fava beans (at 25 °C and L16:D8) that had been supplemented with additional aphids collected across five alfalfa fields 2 weeks prior to the experiment. Dropping propensity can depend on aphid clone (Braendle & Weisser, 2001), so we attempted to use as many clones as possible by taking aphids from across numerous plants from throughout the colony.

We randomly assigned arenas to one of the four treatments and ran 29–34 replicates of each treatment, always using a single predator in each arena and using a new predator, plant, and set of aphids with each replicate. The experiment was run over 11 days with treatments randomly assigned each day based on predator availability. Predators were from recently established laboratory colonies fed with pea aphids, and predators had been satiated then starved for 24 h immediately prior to the start of the experiment to standardize their hunger level and help induce foraging. If a predator did not move within the first 10 min, it was replaced by a predator of equivalent stage. We allowed predators to forage for 2 h, after which we removed the predator and counted the number of aphids remaining on the plant and the number that had dropped. From the original 30 aphids, we subtracted the number left on the plant and the number dropped at the end of the replicate to determine the number of 'missing' aphids. We estimated the number of aphids eaten in all predatory replicates by subtracting the average number missing in replicates of the no predator treatment (mean ± SE: 1.06 ± 0.20) from the observed number of missing aphids. Besides analyzing the estimated number of eaten aphids, we ran all analyses on a second dependent variable, the total number of missing aphids; all analyses yielded the same statistical results, so we do not report the latter.

Two-dimensional arenas

To quantify predator foraging in the absence of aphid dropping, we designed 2-D arenas to replicate the environment of 3-D arenas while preventing aphids from dropping. The 2-D arena used the same cylindrical sleeves and mesh tops and bottoms as used in the 3-D arenas. However, in this arena aphids were restricted to the bottom of the cage on small, excised plant parts rather than being on an upright plant. In the bottom of the arena, we placed four pieces of fava stem (2–3 cm long each) to give aphids a feeding site and account for some potential plant cues. Preliminary observations indicated that most aphids found and fed on the stems and that the amount of aphid movement was similar in 2-D and 3-D arenas. Moreover, we never observed aphids dropping from the small plant parts. Aphids would occasionally walk away or use other defensive behaviors in response to a predator, but they never dropped the 1–2 cm from the stem to the cage bottom. The mesh bottom of the cage was 254.16 cm^2, which was approximately the same surface area as the fava bean plants used in the 3-D arenas. Thus, the surface area available to aphids was kept constant in the two arenas. However, because of the smaller amount of plant material in the 2-D arena, the total amount of surface area available to adult predators (plant + cage) was 9% less in the 2-D arena. This difference should not have affected treatments with larval predators as they cannot fly and they were restricted to the same area as the aphids. Even though we controlled where aphids could be found, the 2-D arenas lacked the structural complexity of the 3-D arenas, which could be a possible confounding factor. Yet, of a large number of potential designs we tested, this design resulted in the most natural behaviors of both predators and aphids while still preventing aphid dropping. We performed the 2-D experiment in the same way as the 3-D experiment, although we omitted the no predator control, because aphid counts were extremely accurate in the 2-D arenas. There were 33–44 replicates of each of the three treatments and the entire experiment was run over 10 days with treatments randomly assigned each day based on predator availability.

Behavioral observations

To better understand how predator foraging behavior may differ among instars and in response to aphid dropping, we made behavioral observations on a subset of 3-D and 2-D replicates using each *H. axyridis* life stage. We observed 9–12 ladybeetles of each life stage in each type of arena (28 in 2-D and 33 in 3-D) and recorded the location and behavior of the predator every minute for the duration of the 2-h replicate. We made observations in 1-min intervals, because preliminary experiments indicated more frequent observations gave the same results. Predator behavior was categorized as (i) moving, (ii) sitting, or (iii) eating (predator mouthparts in contact with an aphid). Location in the 3-D arenas was recorded as either the bottom or top of the specific leaf the predator was on, if it was on the stem, or if it was on the bottom, top, or sides of the cage. Location of foraging ladybeetles was recorded in the 2-D arenas using a 11 × 16 cm grid on the bottom of the cage. We calculated the net displacement of beetles between observations by estimating distances between locations in 3-D arenas or tabulating the distance between grid spaces in 2-D arenas. These measurements were highly correlated with the number of movement observations, so we simply report the latter here.

Statistical analysis

We used four separate analyses of variance (ANOVA) with data from 3-D arenas to determine how predator life stage affected the number of aphids dropped, the number of aphids eaten, the proportion of aphids that dropped that were adult (arcsine square root transformed), and the proportion of aphids that were eaten that were adult (arcsine square root transformed). We used PROC MIXED in SAS (SAS Institute Inc., Cary, NC, USA) for the analysis, since we also wanted to include day as a blocking term, but not all treatments were represented in every block (Littell et al., 1996); we treated block as a fixed effect in the mixed models. We used a similar analysis to determine the number of aphids eaten and the proportion of aphids eaten (arcsine square root transformed) that were adults in 2-D arenas.

The experiments with 2-D and 3-D arenas were originally designed independently, but it became clear that an important and interesting comparison existed between the experiments. Unfortunately, 2-D arenas and 3-D arenas were rarely run at the same time (1 out of 20 possible blocks), and half of the replicates in the 2-D experiment were run before starting the 3-D experiment. Therefore, for analyses that combine data from both experiments, we treated day (block) as a random effect. This assumes that there is a single distribution of variances associated with running the experiment on any given day and that we drew from this distribution each day either experiment was run. This approach is justified, because we saw no temporal trends in any of the data collected in this highly controlled experiment. In addition to the blocking term, this ANOVA included tests for the effects of predator life stage, arena type (2-D vs. 3-D) and their interaction.

Weather conditions, humidity, temperature, and time of day were recorded with each replicate, because they could affect pea aphid dropping behavior (Roitberg et al., 1979; Brodsky & Barlow, 1986; Andrade & Roitberg, 1995; Losey & Denno, 1998a). However, none of these environmental factors appeared to influence the results nor were they
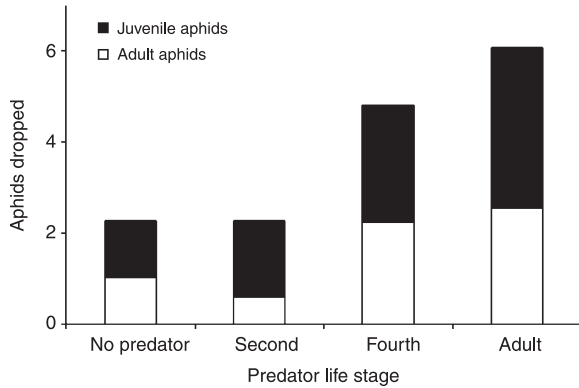
Figure 1 Number of pea aphids, *Acyrthosiphon pisum*, that dropped across predator, *Harmonia axyridis*, life stage treatments in the 3-D experiment with aphid dropping allowed. Columns are divided into the number of adult aphids and juvenile aphids that dropped.

remotely statistically significant in any of our analyses. We used regression analyses to determine how predator movement affected aphid dropping, and how predator movement affected the number of aphids eaten.

Results

Pea aphid dropping rates varied greatly in treatments with different life stages of *H. axyridis* (ANOVA: $F_{3,112} = 12.8$, P<0.001) (Figure 1). The greatest number of aphids dropped in the adult ladybeetle treatment, while fewer dropped in the fourth-instar treatment, and the least dropped in the second-instar treatment. There was no difference in the number of aphids that dropped in the second-instar treatment compared to the number that dropped in the no predator control treatment (Tukey–Kramer: P>0.1).

Juveniles dropped slightly more often than adult pea aphids in the ladybeetle adult, fourth instar, and no predator treatments; on average 58% of aphids that dropped were juveniles. For the second-instar ladybeetle treatment, however, juvenile aphids dropped at a greater rate, as juveniles made up 75% of all dropped aphids. Nonetheless, the overall effect of predator treatment on the proportion of adults that dropped was, at best, marginally significant (ANOVA: $F_{3,105} = 2.08$, P = 0.10).

The number of pea aphids consumed in the 3-D experiment varied by predator life stage (ANOVA: $F_{3,112} = 22.66$, P<0.001) (Figure 2). The greatest number of aphids was eaten in the adult *H. axyridis* treatment, with fewer eaten in the fourth-instar treatment, and the least eaten in the second-instar treatment. In the 2-D experiment, predator life stage was again highly significant and the pattern of aphid predation by life stage was the same as for the 3-D

experiment (ANOVA: $F_{2,99} = 31.31$, P<0.001) (Figure 2); the number of aphids consumed decreased successively from adults to fourth to second instars.

In all cases, a given stage of ladybeetle preyed upon more aphids in the 2-D experiment than in the 3-D experiment; thus, ladybeetles consume fewer aphids when aphids are allowed to drop (Figure 2). Since the pattern of predation between life stages was the same in both 2-D and 3-D experiments, there was no interaction between predator life stage and type of arena on the number of aphids consumed ($F_{2,180} = 0.17$, P = 0.840). As expected from the results above, predator treatment was highly significant ($F_{2,180} = 36.5$, P<0.001), and because predation was consistently lower for each predator life stage when run in a 3-D arena rather than a 2-D arena, the term for type of arena was also highly significant ($F_{2,202} = 37.2$, P<0.001).

Predator treatment did not affect the proportion of juvenile vs. adult aphids that were eaten in either the 3-D arenas ($F_{3,94} = 1.35$, P = 0.262) or 2-D arenas ($F_{2,99} = 0.09$, P = 0.913) when each was analyzed independently. When analyzed together, there was no effect of predator treatment, and no interaction effect between predator treatment and arena type (P>0.1). However, the proportion of juvenile vs. adult aphids eaten differed between 2-D and 3-D treatments. In the 2-D experiment, 45% of the aphids eaten were adults, while 59% of those eaten in the 3-D experiment were adults (ANOVA: $F_{1,176} = 5.87$, P = 0.016). These results were not due to observer bias in reporting one aphid stage 'missing' more often in the 3-D experiment; the mean proportion of adults seen missing in the no predator treatment was not different than 0.5 (Wilcoxon sign test, P>0.1).
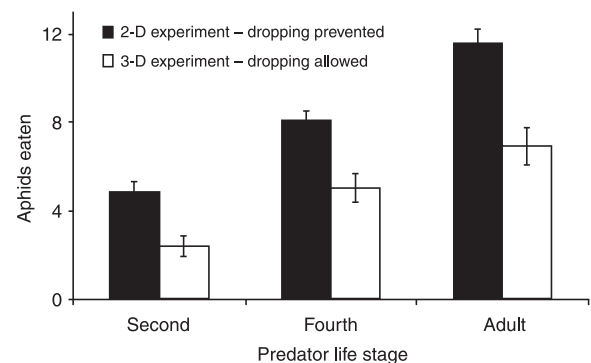
Figure 2 Number of pea aphids, *Acyrthosiphon pisum*, eaten (mean ± SE) by each predator, *Harmonia axyridis*, life stage in the 2-D experiment (no aphid dropping) and 3-D experiment (aphid dropping allowed). Predation was significantly greater for each predator treatment in the 2-D experiment.
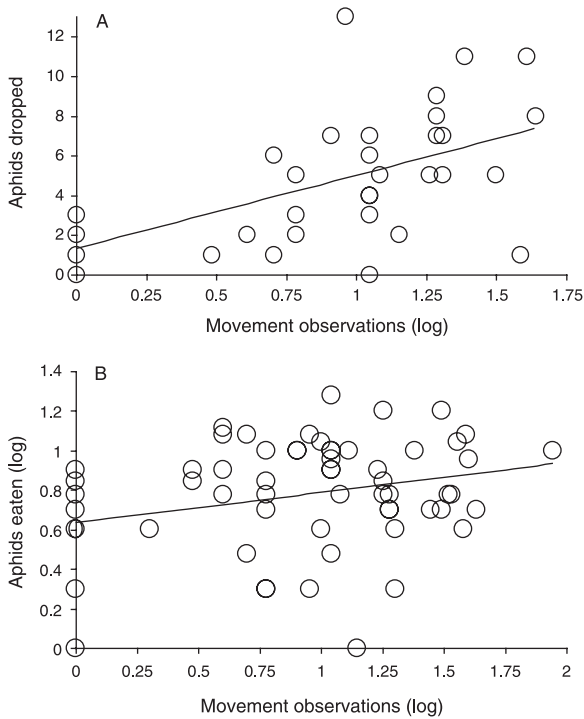
Figure 3 (A) Relationship between the number of pea aphids, *Acyrthosiphon pisum*, that dropped in a replicate and the number of movement observations recorded for the predator, *Harmonia axyridis*, in that replicate (3-D experiment only) ($r^2 = 0.293$, $P = 0.011$). (B) Relationship between the number of aphids eaten in a replicate and the number of movement observations recorded for the predator in that replicate (both experiments) ($r^2 = 0.077$, $P = 0.013$).

Behavioral observations

Observations of predator behavior, across all life stages, showed that more aphids dropped when predator movement increased (regression: $F_{1,29} = 7.3$, $P = 0.011$) (Figure 3A). Likewise, across all life stages and replicates, as predator movement increased, the number of aphids eaten increased (regression: $F_{1,56} = 6.5$, $P = 0.013$) (Figure 3B).

Discussion

In both arenas where pea aphids were and were not allowed to drop, pea aphid predation increased with older predator life stages (Figure 1). By comparing the results from these arenas, we see that pea aphid predation is greatly reduced when pea aphids are allowed to drop (Figure 2). This suggests that the foraging of *H. axyridis* induces pea aphids to drop from plants, and that this dropping behavior reduces the foraging efficiency of *H. axyridis*. Despite the differential predation on aphids among ladybeetle life stages, dropping behavior reduced predation on aphids by

about 40% for all predator life stages (Figure 2). Aphid dropping and ladybeetle foraging efficiency were both correlated with predator movement, which also tended to increase with older predators.

It is important to consider how much of the difference between experiments was due to aphid dropping behavior and how much may be due to other differences caused by the artificial arenas. Despite controlling for the surface area where prey could be found, the total surface area available for an adult ladybeetle to forage upon was 9% greater in the 3-D arena than in the 2-D arena. This may make it harder for the ladybeetle to find the prey and reduce its efficiency in the 3-D arena. However, because the flightless ladybeetle larvae were restricted to the same area that aphids were, the total surface area they could forage upon was the same in both 2-D and 3-D arenas. If the small difference in total surface area was an important factor, we would have expected that the aphid predation differential between arenas to be larger for adults than it was for other life stages. Since the proportional effect was the same for all ladybeetle life stages, total surface area was not likely to have been important. The 2-D and 3-D arenas were also different in that the 2-D arena was much more artificial than the 3-D arena that was centered on an excised plant infested with aphids. If ladybeetles are not as effective at foraging on mesh as they are on plant materials (due to differences in tactile or aphid cues), we would expect lower predation rates in the more artificial 2-D arena. If that is the case, our estimate of the effect of dropping is a conservative one. The other major difference in arenas relates to surface topology; despite equal surface areas, the plant is structurally more complex than the mostly flat mesh bottom. Such differences in structural heterogeneity can affect predator movement and foraging behavior (reviewed in Cortesero et al., 2000), though not necessarily in a consistent or straightforward manner (e.g., Grez & Villagran, 2000).

Because we did not run all replicates of each experiment at the same time, our experimental design did not have balanced blocks. However, there were no temporal trends in any of our results, and we accounted for the unbalanced blocking design experiment by treating block as a random effect. Moreover, all replicates were performed within a 3-month period, using same-aged ladybeetles (including adults) and using aphids from a large, clonally diverse colony. Therefore, our results are statistically valid despite the unbalanced blocking design.

Losey & Denno (1998a) concluded that the escape response of a pea aphid should be positively associated with predator size and, therefore, its apparent risk to aphids. Our results matched this prediction: aphids dropped more frequently with increasing predator stage in the 3-D experiment. When the predator is large, an aphid's most

effective defense mechanism may be dropping, whereas an aphid may be able to survive a smaller predator's attack by performing less costly behaviors, such as walking away, kicking (Roitberg & Myers, 1978; Brodsky & Barlow, 1986), or fending off predators with their cornicles (Mondor & Roitberg, 2002).

The behavior of the predator, in particular its movement rate, appears to play a large role in aphid dropping (Figure 3). This may be because predator movement causes vibrations in the plant, which aphids can detect and use as a cue to drop (Brodsky & Barlow, 1986; Hajek & Dahlsten, 1987; Losey & Denno, 1998a). If greater movement rates cause more vibrations, this could explain the observed correlation between predator movement and aphid dropping. Predator movement may also correlate with aphid encounters. More encounters could directly lead to more predation (suggested by Figure 3B) as well as more dropping (suggested by Figure 3A). Increased encounters could also result in more aphids secreting alarm pheromone, triggering other aphids nearby to drop (Mondor & Roitberg, 2000). Moreover, alarm pheromone sometimes gets smeared on the predator itself (Mondor & Roitberg, 2004), thereby providing an additional way that increased predator movement could increase aphid dropping.

Previous studies have shown that adult aphids have a greater tendency to drop from plants than juvenile aphids (Roitberg & Myers, 1979; McConnell & Kring, 1990; Losey & Denno, 1998a,b; Mondor & Roitberg, 2002). This is presumably because adults walk faster and can more easily find a new host plant before desiccating or encountering ground-foraging predators (Gish & Inbar, 2006). However, in our study adult aphids did not drop more than juveniles; in most treatments, slightly more juveniles dropped than adults. In our second-instar predator treatment, however, a much greater number of juvenile aphids dropped than adult aphids (paired t-test: t_{28} = 3.88, P<0.001). This may be because small ladybeetle larvae are hunting more for smaller aphids or because adult aphids do not sense the young ladybeetles to be much of a threat.

While aphid dropping behavior reduces the amount of direct predation, to measure the total impact of *H. axyridis*, we would have to combine direct effects of predation (as above) with additional indirect and sublethal effects that come about because of dropping. Aphids that drop can incur 6–33% mortality due to desiccation depending upon climactic conditions (Roitberg & Myers, 1978). Those that survive exposure to high temperatures can still have substantially lower fecundity depending upon the aphid's age and compliment of secondary bacterial symbionts (e.g., Russell & Moran, 2006). Dropping can also create risks from ground-foraging predators, such as the carabids that consumed 64% of the aphids that had escaped a foliar-dwelling predator (Losey & Denno, 1998c). Even in cases where there are few other predators and climactic conditions are favorable, foraging by natural enemies can negatively impact aphid reproduction through sublethal effects (Tamaki et al., 1970). Field estimates suggest that a single predator encounter can cost an aphid an hour in feeding time and result in a 7.6% reduction in reproductive output (Nelson, 2007). Therefore, in the most extreme circumstances, we can estimate that the total impact of *H. axyridis* on pea aphids could be almost double the direct effect it has via predation.

Aphid dropping behavior reduced predator foraging in our simple laboratory environments and could be similarly important in influencing *H. axyridis* and its usefulness as a biological control agent across agroecosystems. In the mid-western USA, two of the most abundant prey for *H. axyridis* are pea aphids in alfalfa fields and soybean aphids, *Aphis glycines*, in soybeans. These aphids are both good food sources for *H. axyridis*; they are nutritionally similar for developing ladybeetle larvae (KJ Forbes, unpubl.) and induce identical oviposition responses in gravid *H. axyridis* females (PP Lee, unpubl.). Nonetheless, *H. axyridis* is commonly found in much greater densities in soybean fields than in alfalfa fields (KJ Forbes, unpubl.). One of the important differences between these systems is that soybean aphids, unlike pea aphids, do not exhibit dropping behavior. This may be particularly important for *H. axyridis*, which seems to tightly aggregate to high densities of aphids at a small spatial scale (e.g., Cardinale et al., 2006). Thus, variation in antipredator behaviors may be a potentially important reason for the variation in this generalist predator's effectiveness as a biological control agent in different systems.

Acknowledgements

References

Andrade MCB & Roitberg BD (1995) Rapid response to intraclonal selection in the pea aphid (*Acyrthosiphon pisum*). Evolutionary Ecology 9: 397–410.

Braendle C & Weisser WW (2001) Variation in escape behavior of red and green clones of the pea aphid. Journal of Insect Behavior 14: 497–509.

Brodsky LM & Barlow CA (1986) Escape responses of the pea aphid, *Acyrthosiphon pisum* (Harris) (Homoptera: Aphididae): influence of predator type and temperature. Canadian Journal of Zoology 64: 937–939.

Cardinale BJ, Weis JJ, Forbes AE, Tilmon KJ & Ives AR (2006) Biodiversity as both a cause and consequence of resource availability: a study of reciprocal causality in a predator-prey system. Journal of Animal Ecology 75: 497–505.

Chau A & Mackauer M (1997) Dropping of pea aphids from feeding site: a consequence of parasitism by the wasp, *Monoctonus paulensis*. Entomologia Experimentalis et Applicata 83: 247–252.

Cortesero AM, Stapel JO & Lewis WJ (2000) Understanding and manipulating plant attributes to enhance biological control. Biological Control 17: 35–49.

Dill LM, Fraser AHG & Roitberg BD (1990) The economics of escape behavior in the pea aphid, *Acyrthosiphon pisum*. Oecologia 83: 473–478.

Gish M & Inbar M (2006) Host location by apterous aphids after escape dropping from the plant. Journal of Insect Behavior 19: 143–153.

Grez AA & Villagran P (2000) Effects of structural heterogeneity of a laboratory arena on the movement patterns of adult *Eriopis connexa* and *Hippodamia variegata* (Coleoptera: Coccinellidae). European Journal of Entomology 97: 563–566.

Gross P (1993) Insect behavioral and morphological defenses against parasitoids. Annual Review of Entomology 38: 251–273.

Hajek AE & Dahlsten DL (1987) Behavioral interactions between three birch aphid species and *Adalia bipunctata* larvae. Entomologia Experimentalis et Applicata 45: 81–87.

Honěk A, Jarosik V, Lapchin L & Rabasse JM (1998) The effect of parasitism by *Aphelinus abdominalis* and drought on the walking movement of aphids. Entomologia Experimentalis et Applicata 87: 191–200.

Koch RL (2003) The multicolored Asian lady beetle, *Harmonia axyridis*: a review of its biology, uses in biological control, and non-target impacts. Journal of Insect Science 3: 32.

Kunert G, Otto S, Röse USR, Gershenzon J & Weisser WW (2005) Alarm pheromone mediates production of winged dispersal morphs in aphids. Ecology Letters 8: 596–603.

Littell RC, Milliken GA, Stroup WW & Wolfinger RD (1996) SAS® System for Mixed Models. SAS Institute Inc., Cary, NC, USA.

Losey JE & Denno RF (1998a) The escape response of pea aphids to foliar-foraging predators: factors affecting dropping behaviour. Ecological Entomology 23: 53–61.

Losey JE & Denno RF (1998b) Interspecific variation in the escape responses of aphids: effect on risk of predation from foliar-foraging and ground-foraging predators. Oecologia 115: 245–252.

Losey JE & Denno RF (1998c) Positive predator-predator inter-actions: enhanced predation rates and synergistic suppression of aphid populations. Ecology 79: 2143–2152.

McConnell JA & Kring TJ (1990) Predation and dislodgment of *Schizaphis graminum* (Homoptera: Aphididae), by adult *Coccinella septempunctata* (Coleoptera: Coccinellidae). Environmental Entomology 19: 1798–1802.

Mondor EB & Roitberg BD (2000) Has the attraction of predatory coccinellids to cornicle droplets constrained aphid alarm signaling behavior? Journal of Insect Behavior 13: 321–329.

Mondor EB & Roitberg BD (2002) Pea aphid, *Acyrthosiphon pisum*, cornicle ontogeny as an adaptation to differential predation risk. Canadian Journal of Zoology 80: 2131–2136.

Mondor EB & Roitberg BD (2004) Inclusive fitness benefits of scent-marking predators. Proceedings of the Royal Society of London Series B – Biological Sciences 271: S341–S343.

Nelson EH (2007) Predator avoidance behavior in the pea aphid: costs, frequency, and population consequences. Oecologia 151: 22–32.

Nelson EH & Rosenheim JA (2006) Encounters between aphids and their predators: the relative frequencies of disturbance and consumption. Entomologia Experimentalis et Applicata 118: 211–219.

Roitberg BD & Myers JH (1978) Adaptation of alarm pheromone responses of the pea aphid *Acyrthosiphon pisum* (Harris). Canadian Journal of Zoology 56: 103–108.

Roitberg BD & Myers JH (1979) Behavioural and physiological adaptations of pea aphids (Homoptera: Aphididae) to high ground temperatures and predator disturbance. Canadian Entomologist 111: 515–519.

Roitberg BD, Myers JH & Frazer BD (1979) The influence of predators on the movement of apterous pea aphids between plants. Journal of Animal Ecology 48: 111–122.

Russell JA & Moran NA (2006) Costs and benefits of symbiont infection in aphids: variation among symbionts and across temperatures. Proceedings of the Royal Society B – Biological Sciences 273: 603–610.

Snyder WE & Ives AR (2003) Interactions between specialist and generalist natural enemies: parasitoids, predators, and pea aphid biocontrol. Ecology 84: 91–107.

Tamaki G, Halfhill JE & Hathaway DO (1970) Dispersal and reduction of colonies of pea aphids by *Aphidius smithi* (Hymenoptera: Aphidiidae). Annals of the Entomological Society of America 63: 973–980.

Ueno H (2003) Genetic variation in larval period and pupal mass in an aphidophagous ladybird beetle (*Harmonia axyridis*) reared in different environments. Entomologia Experimentalis et Applicata 106: 211–218.

Villagra CA, Ramirez CC & Niemeyer HM (2002) Antipredator responses of aphids to parasitoids change as a function of aphid physiological state. Animal Behaviour 64: 677–683.