

# Comparative biology and pesticide susceptibility of *Amblydromella caudiglans* and *Galendromus occidentalis* as spider mite predators in apple orchards

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Abstract The successful integrated mite management program for Washington apples was based on conservation of the mite predator Galendromus occidentalis (Nesbitt). In the 1960s, this mite was assumed to be the only phytoseiid in Washington commercial apple orchards, due to its preference for the most common mite pest of that period, Tetranychus mcdanieli McGregor, as well as its resistance to organophosphate pesticides. A recent survey of phytoseiids in Washington apple found that another phytoseiid, Amblydromella caudiglans (Schuster) has become common. It is a more generalized predator than G. occidentalis (it is not a Tetranychus spp. specialist) and is not known to be organophosphate-resistant. A series of experiments was conducted to compare the life history, prey consumption, and pesticide tolerance of these two species. Galendromus occidentalis developed more quickly than A. caudiglans, but had slightly lower egg survival. Although A. caudiglans attacked more Tetranychus urticae Koch eggs than G. occidentalis, it could not reproduce on this diet. Both predators performed equally well on a diet of T. urticae protonymphs. Unlike G. occidentalis, A. caudiglans experienced significant mortality when exposed to carbaryl, azinphosmethyl, and bifenazate. Both predators experienced significant mortality due to imidacloprid and spinetoram. These results highlight the key differences between these two predators; the shift away from organophosphate use as well as the change in dominant mite pest to Panonychus ulmi (Koch) may be driving factors for the observed increased abundance of A. caudiglans in Washington apple.

**Keywords** Phytoseiidae · *Galendromus occidentalis · Amblydromella caudiglans ·* Biological control · Spider mite · Apple

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# Introduction

Integrated mite management (IMM) in Washington apple orchards was developed in the 1960s with conservation of *Galendromus occidentalis* (Nesbitt) as the cornerstone (Hoyt 1969; Hoy 2011). This predatory mite was capable of controlling the most abundant spider mite pest, *Tetranychus mcdanieli* McGregor (Hoyt 1969). It had also developed resistance to some of the insecticides commonly used for codling moth (*Cydia pomonella* L.) control, including the organophosphate (OP) azinphosmethyl (Hoyt 1969; Croft and Jeppson 1970; Ahlstrom and Rock 1973; McMurtry 1982). This allowed for the selective use of pesticides for control of key pests while minimizing the disruption of the biological control provided by *G. occidentalis* (Hoy 2011).

However, pest management practices in Washington apple have changed substantially in the intervening decades. Mating disruption of codling moth was registered in 1990, and use in Washington apple increased steadily over the next decade. OP use has been gradually phased out due to loss of efficacy and federal regulations; as of 2013, azinphosmethyl (an OP widely used for codling moth control) can no longer be applied on apples (Environmental Protection Agency 2012). This has been part of a larger movement away from older, broad-spectrum pesticides towards the use of reduced risk/OP replacement pesticides from newer chemistry classes. Associated with these changes in IPM practices, a shift in the most common mite pest species has occurred, with *Panonychus ulmi* (Koch) outbreaks more commonly reported and *T. mcdanieli* outbreaks becoming increasingly rare (Beers and Hoyt 2007).

Because of these changes, a survey of phytoseiid fauna in commercial apple orchards was conducted in Washington from 2011 to 2013 (Schmidt-Jeffris et al. 2015). The survey results found several other phytoseiid species in addition to *G. occidentalis*. Most notably, *Amblydromella caudiglans* (Schuster) was present in 50 % of orchards sampled and was the dominant species in nearly 20 % of orchards. Higher abundances of this species were correlated with weedy herbicide strips and absence of bifenazate use, whereas *G. occidentalis* populations were higher in conventional (vs. organic) orchards and where bifenazate had been used (Schmidt-Jeffris et al. 2015). These results indicate that tolerance of disturbance by certain pesticides could be the key difference between these predators.

The two phytoseiids studied are also known to have different dietary preferences. *Galendromus occidentalis* is a specialist on spider mites in the genus *Tetranychus*; it is attracted to volatiles produced by mites while feeding and is capable of navigating the copious amounts of webbing that *Tetranychus* spp. produces (Sabelis and Van de Baan 1983; McMurtry and Croft 1997). However, it can also feed on other tetranychid species (including *P. ulmi*) as well as eriophyids, such as *Aculus schlechtendali* (Nalepa) (Hoyt and Beers 1993). This places *G. occidentalis* in the "Type II" specialist category, indicating that it is loosely specialized on tetranychids, but is commonly associated with web-producing *Tetranychus* spp. (McMurtry and Croft 1997; McMurtry et al. 2013).

Alternatively, A. caudiglans prefers to feed on eriophyids or spider mites outside of the genus *Tetranychus*, and has difficulty moving in the web-nests produced by *Tetranychus* spp. (Putman 1962; Clements and Harmsen 1993; Blackwood et al. 2004). Unlike *G. occidentalis*, it can also reproduce while feeding exclusively on pollen (Putman 1962; Blackwood et al. 2004). This species is placed in the "Type III" category (McMurtry and Croft 1997), or "Type III-a" (McMurtry et al. 2013). This category indicates that it is more generalized than Type II species (it can reproduce on pollen, is not associated with *Tetranychus* spp.) and further indicates its preferred habitat type "a"—pubescent leaves. Like

*G. occidentalis*, this phytoseiid can also reproduce on *P. ulmi* (Putman 1962). It has also been suggested that *A. caudiglans* is the primary control agent of *P. ulmi* in Ontario peach orchards (Putman and Herne 1966). Therefore, a shift away from *T. mcdanieli* to *P. ulmi* as the most common pest species could also favor higher densities of *A. caudiglans*.

The series of studies described here were conducted with the purpose of better describing the differences between these two common orchard phytoseiids. A series of assays was conducted to determine how these species differ in terms of life stage duration, survival, prey consumption and fecundity on two prey classes (eggs and protonymphs), and pesticide resistance. These experiments serve to provide additional clarification as to why a given phytoseiid species may be abundant in one orchard, but not another.

### Materials and methods

### Life history

*Amblydromella caudiglans* were obtained from an unsprayed research orchard in Wenatchee, WA. Adult females were placed individually on lima bean (*Phaseolus vulgaris* L. 'Henderson Bush') leaf disk arenas and monitored for oviposition every 12 h. When an egg was laid, the female was slide-mounted and identified to species following the key of Denmark and Evans (2011); if the female was *A. caudiglans*, the egg was then used for the life table study. *Galendromus occidentalis* individuals were obtained from a colony established from a field population collected from a research orchard near Orondo, WA in June 2013. The research orchard had a history of being minimally sprayed; only pyriproxifen (Esteem 35WP, Valent USA, Walnut Creek, CA, USA) was used for codling moth control. This colony was maintained on *Tetranychus urticae* Koch kept on lima bean plants. Individual phytoseiids from the colony were slide-mounted to confirm species identity, and the colony was checked before use to ensure contamination by other species had not occurred. Females from the *G. occidentalis* colony were placed individually on leaf disk arenas until oviposition. When an egg was laid, it was used in the life table study.

Leaf disk arenas were constructed by cutting a 2.2 cm diameter disk from a lima bean leaf. A plastic cup (14.7 mL) was filled half-way with agar (Bacto<sup>TM</sup> Agar, BD, Sparks, MD, USA). When the agar had cooled, but not completely solidified, the bean leaf disks were placed into the agar with the abaxial surface facing up. The outer edges of the disks were gently pressed into the agar to hold the leaf flat and to maintain leaf turgor. A band of adhesive material (Tangle-Trap Insect Trap Coating, The Tanglefoot Company, Grand Rapids, MI, USA) was applied in a ca. 1 cm band near the inside upper edge to prevent escape. A small tuft of cotton was adhered to the leaf disk with agar to provide the female mite with shelter. The bioassay cups were sealed with friction-fit lids in which a 1 cm diameter hole was cut and covered with surgical tape (Micropore, 3 M, St. Paul, MN, USA) for ventilation while not allowing escape.

Individual eggs were monitored in these arenas, with one egg per arena. A sufficient number of individuals (replicates) were monitored so that at least n = 50 individuals of each species reached adulthood. Upon hatch, juveniles were provided with *T. urticae* in various stages, ad libitum. Apple pollen was also brushed onto all leaf disks (regardless of species) to provide a secondary food source for *A. caudiglans*. After hatching, the life stage and condition (alive or dead) of the mite was recorded every 12 h. Stages were distinguished by the number of legs (larvae vs. protonymphs) and size. Arenas were held in a

growth room at 24–32 °C, 30–50 % relative humidity with a 16:8 h day length, conditions which simulated mid-summer conditions in central Washington. Each individual (from an egg) constituted a single replication.

Life stage durations for each species were compared using PROC GLIMMIX, specifying the negative binomial distribution in the model statement; the model *F* test was used to compare the two species ( $\alpha = 0.05$ ). Survival and sex ratio were compared using a logistic regression model (PROC GENMOD) using a logit link. The variables (dead/live, male/female) were treated as binomial, with the binomial distribution specified in the model statement; Wald  $\chi^2$  tests were used to compare the two species ( $\alpha = 0.05$ ).

### Prey consumption (eggs) and fecundity

The arena for the bioassays consisted of a 2.2 cm diameter bean leaf disk cut from an untreated, uninfested bean leaf, and placed with the abaxial surface facing up in a plastic cup (14.7 mL) filled with cotton and water. Thirty *T. urticae* females were added to each arena and allowed to oviposit for 24 h. Adult females were removed from the disks, and egg numbers were adjusted to 40 per disk and the position of each egg was marked with a felt-tip pen. One adult female phytoseiid was added to each of 30 disks (replicates). *Galendromus occidentalis* were obtained from a research orchard in Orondo, WA, and *A. caudiglans* were obtained from apple trees in a research orchard in Wapato, WA the day prior to loading; previous experiments indicated that relative treatment differences (the rank of each pesticide from least to most toxic) were maintained with field-collected predators compared to their counterparts from synchronous cohorts (Beers et al. 2009). The Orondo orchard spray regime was as previously described. The Wapato orchard had been treated with chlorantraniliprole (Altacor 35WDG, E.I DuPont de Nemours & Co., Wilmington, DE, USA) and spinosad (Entrust, Dow AgroSciences, Indianapolis, IN, USA) during the year of collection. Females collected from these orchards were identified to species as described above.

Remaining *T. urticae* eggs were counted after 48 h. Arenas were held an additional 48 h to allow the phytoseiids to oviposit. At the end of this period, all phytoseiid eggs and larvae (some eggs had hatched) were counted.

#### Prey consumption (protonymphs) and fecundity

A second experiment, using *T. urticae* protonymphs instead of eggs, was conducted in a similar manner. However, 20 protonymphs from a *T. urticae* colony were transferred to the arena, and phytoseiid females were added immediately after. Live and dead *T. urticae* protonymphs were recorded at the end of the 48 h period; oviposition of the phytoseiids was counted 48 h after that evaluation.

Data from both prey consumption/fecundity experiments were analyzed using PROC GLIMMIX (SAS 2014), specifying the negative binomial distribution for count data; the model *F* test was used to compare the two species ( $\alpha = 0.05$ ). Replicates where the phytoseiid could not be found at the end of the study were excluded from the analysis.

#### Pesticide toxicity

The arena for the bioassays consisted of a 2.2 cm diameter bean leaf disk cut from an untreated, uninfested bean leaf, and placed with the abaxial surface facing up in a plastic cup (14.7 mL) filled with cotton and water. A sufficient number of mixed stages of *T*.

*urticae* to feed predators were added to each arena by brushing prey from infested bean leaves using a 3 cm paint brush. A small quantity of apple pollen and a single phytoseiid female was also added to each leaf disk. These mites were obtained directly from apple leaves collected the previous day from research orchards in Rock Island, WA (*G. occidentalis*) and Wapato, WA (*A. caudiglans*). The research orchard in Rock Island had a typical thinning and codling moth program, consisting of the materials chlorantraniliprole, methoxyfenozide (Intrepid 2F, Dow AgroSciences), acetamiprid (Assail 70WP, United Phosphorus, King of Prussia, PA, USA), and spinetoram (Delegate 25WG, Dow AgroSciences). The pesticide regime at the Wapato orchard was as previously described. A total of 25 arenas (replicates) were used for each treatment.

The treatments were applied as a topical spray to the phytoseiid females on the disks, and they remained on the same disk throughout the bioassay period. Thus exposure to the pesticide combined contact, residues, and contaminated prey. The pesticide concentration used was based on the maximum label rate of the pesticide per unit surface area applied in 935 L/ha of water. The solutions were made by mixing the appropriate amount of the formulated pesticide in 1 L of water. Pesticides were applied with a laboratory sprayer (Potter Spray Tower, Burkard, Rickmansworth, UK) set at 44.8 kPa using the intermediate nozzle. Each arena was sprayed with 2 mL of pesticide mixture (deposition at these settings was 1.94 mg solution/cm<sup>2</sup>); the checks were sprayed with distilled water.

The pesticides tested were those commonly used in eastern Washington apple production, and represented a wide range of modes of action (Table 1). Some were included because they were indicated as potentially important factors affecting phytoseiid

Common name	Mode of action <sup>a</sup>	Chemical class	Brand name/ formulation <sup>b</sup>	Formulation	Use rate (g ai/ ha) <sup>c</sup>	mg AI/l (bioassay rate)
Carbaryl	1A	Carbamate	Sevin 4F <sup>d</sup>	479 g/l	3363	3595
Azinphosmethyl	1B	Organophosphate	Guthion 50W <sup>d</sup>	500 g/kg	1681	1798
Imidacloprid	4A	Neonicotinyl	Provado 1.6F <sup>d</sup>	192 g/l	112	120
Spinetoram	5	Spinosyn	Delegate 25WG <sup>e</sup>	250 g/kg	123	131
Novaluron	15	IGR—benzoyl urea	Rimon 0.83EC <sup>f</sup>	99 g/l	363	389
Spirotetramat	23	Tetramic acid	Ultor 1.25L <sup>d</sup>	150 g/l	153	164
Chlorantraniliprole	28	Anthranilic diamide	Altacor 35WDG <sup>g</sup>	350 g/kg	110	118
Bifenazate	UN	-	Acramite 50WS <sup>f</sup>	500 g/kg	1121	599

 Table 1
 Pesticides used in nontarget effects bioassays

<sup>a</sup> Mode of action classification taken from Insecticide Resistance Action Committee (IRAC) v 7.3 (http:// www.irac-online.org/documents/moa-classification/?ext=pdf) or the fungicide Resistance Action Committee (FRAC)

<sup>b</sup> The registrant listed is from the time the experiments were begun

- <sup>c</sup> The concentrations were based on an application rate of 935 l/ha, or 100 US gallons/acre
- <sup>d</sup> Bayer CropScience, Research Triangle Park, NC, USA
- <sup>e</sup> Dow Agrosciences, Indianapolis, IN, USA
- <sup>f</sup> Chemtura Corporation, Middlebury, CT, USA

<sup>&</sup>lt;sup>g</sup> E.I DuPont de Nemours & Co., Wilmington, DE, USA

abundance (Schmidt-Jeffris et al. 2015). Numbers of live, dead, and runoff phytoseiid females were recorded 48 h after treatment. Arenas were held at  $20 \pm 2^{\circ}$  C and 16:8 L:D photoperiod.

Data from the female bioassays were analyzed using a logistic regression model (PROC GENMOD, SAS 9.3 (SAS Institute, 2013)) using a logit link. Mortality (dead + runoff) and runoff (alone) were treated as binomial (live/dead, runoff/live), with the binomial distribution specified in the model statement. Pesticides within a species were compared when the overall model was significant ( $\alpha = 0.05$ ) using pairwise single degree-of-freedom likelihood ratio contrasts ( $\alpha = 0.05$ ).

## Pesticide repellency

Pesticides with high levels of runoff (irritancy) in the mortality bioassays were further tested for repellency. Repellency is defined as the avoidance of treated surfaces by remaining on untreated surfaces, whereas irritancy is escaping residues entirely by running off of the disk (Beers and Schmidt-Jeffris 2015). Only *A. caudiglans* was tested; *G. occidentalis* had already been screened in previous work (Beers and Schmidt-Jeffris 2015). The bioassay arena consisted of a bean leaf disk 3.5 cm in diameter. The disk was cut so that it was bisected by the midvein, which served as the division between the treated and untreated halves. The disk halves were treated by dipping them for 3 s in the appropriate pesticide; the other half was left untreated. After treatment with a pesticide, the disk was allowed to dry for ca. 1 h, and then was placed with the abaxial surface facing up in a plastic cup (30 mL) filled with cotton and water. The pesticide treatment was replicated five times.

Ten *A. caudiglans* females were transferred to each disk, placing them on the midvein to avoid bias. The females were allowed to settle for ca. 2 h, then the numbers of live and dead mites on the treated versus the untreated leaf half were recorded. This evaluation was repeated two more times at ca. 2 h intervals, for a total of three evaluations. Runoff (leaving the disk arena) was determined by subtraction, and mortality was a composite of all mites found dead, regardless of which side they were found on. Between evaluations, arenas were held at  $20 \pm 2^{\circ}$  C.

Repellency was assessed with logistic regression using PROC GENMOD. Each pesticide was tested and analyzed independently. Within each pesticide the proportion at each evaluation was assessed separately using the Wald test. The research hypothesis for each of these tests was that the true underlying proportion deviated from 0.5 (50 %) or equivalently, the log-odds ratio deviated from 0. Significance was declared at P < 0.0167, using the Bonferroni adjustment for three comparisons (three evaluations) to the 5 % level of significance.

## **Results and discussion**

## Life history

*Galendromus occidentalis* had a shorter egg to adult development time than *A. caudiglans* (Table 2), but the difference was only 0.59 days. Although *A. caudiglans* had shorter egg and larval stages, *G. occidentalis* had shorter nymphal stages. Unlike *G. occidentalis*, *A.* 

Species	Duration	ı in days								
	u	Egg	и	Larva	и	Protonymph	и	Deutonymph	и	Total
A. caudiglans	107	$1.94 \pm 0.04$	95	$0.65\pm0.03$	67	$1.64 \pm 0.07$	50	$1.91 \pm 0.08$	50	$5.94 \pm 0.14$
G. occidentalis	113	$2.16\pm0.04$	91	$0.99\pm0.03$	84	$1.05\pm0.06$	76	$1.19 \pm 0.07$	76	$5.35\pm0.12$
$df_1$ , $df_2$		1, 218		1, 184		1, 149		1, 124		1, 124
F		18.74		58.49		44.40		44.29		10.20
Ρ		<0.01		<0.01		<0.01		<0.01		<0.01

*caudiglans* larvae do not feed (Putman 1962); non-feeding larvae are known to develop more quickly (Schausberger and Croft 1999). *Amblydromella caudiglans* had higher survival in the egg stage than *G. occidentalis*, but survival in all other stages was similar (Table 3). Both *A. caudiglans* and *G. occidentalis* had similar sex ratios, 74 and 62 % female, respectively (Table 3). Although the small differences in life stage duration and survival were statistically significant, it is unlikely that they will be biologically significant in terms of predator performance. It is understood that these results cannot be extrapolated beyond the populations tested, but these populations are likely representative.

### Prey consumption (eggs) and fecundity

*Amblydromella caudiglans* consumed more *T. urticae* eggs than *G. occidentalis* (Table 4). However, this diet resulted in low fecundity for *A. caudiglans* (Table 4). Previous research has suggested that *T. urticae* eggs are a poor source of nutrition for *A. caudiglans*, which could explain the reduced oviposition (Putman 1962). Putman (1962) also suggested that *A. caudiglans* might be puncturing and draining eggs, but not necessarily consuming their contents. Generalist phytoseiids feeding on *T. urticae* are known to prefer motile life stages over eggs (Blackwood et al. 2001).

## Prey consumption (protonymphs) and fecundity

Both phytoseiids consumed the same number of prey when fed a diet of *T. urticae* protonymphs (Table 4). Both species also laid a similar number of eggs on this diet. The over  $4 \times$  increase in fecundity of *A. caudiglans* on this diet, as opposed to the eggs-only diet, supports the hypothesis that the egg diet was nutritionally inadequate. However, because these two studies were not conducted simultaneously, they cannot be compared statistically.

The fecundity for *G. occidentalis* for this (and the previous) study was quite low compared to previously published studies (Lefebvre et al. 2011; Beers and Schmidt 2014). This may be a characteristic of this particular population. However, an increase in fecundity would only accentuate the better performance of *G. occidentalis* on *T. urticae*, compared to *A. caudiglans*.

Because A. caudiglans is less capable of reproducing on one stage (eggs) of T. urticae, this may have limited its ability to reach high abundances when *Tetranychus* spp. were more common pests of orchards than they are at present (Beers and Hoyt 1993). Reduced fecundity on this prey may be a contributing factor to the previous dominance of G.

Species % survival									п	% female
	n	Egg	п	Larva	n	Protonymph	п	Deutonymph		
A. caudiglans	109	98.17	104	95.58	73	91.78	55	90.91	55	74.14
G. occidentalis	131	86.26	103	89.32	88	95.45	76	96.20	76	61.84
$\chi^2 (df = 1)$		12.87		2.56		0.92		1.59		2.04
Р		< 0.01		0.11		0.34		0.21		0.15

 Table 3
 Stage-specific survival and sex ratio of Amblydromella caudiglans and Galendromus occidentalis

 fed a diet of apple pollen and mixed stages of Tetranychus urticae

	Tetranychus urticae eş	ggs	T. urticae protonymphs				
	Prey consumed/48 h	Eggs laid/96 h	Prey consumed/48 h	Eggs laid/96 h			
A. caudiglans	28.17 (21.72, 36.54)	0.37 (0.18, 0.74)	9.80 (8.35, 11.51)	1.67 (1.22, 2.27)			
G. occidentalis	8.99 (6.54, 12.36)	1.57 (1.05, 2.35)	10.23 (8.73, 11.99)	1.33 (0.94, 1.87)			
$df_1, df_2$	1, 17	1, 24	1, 29	1, 29			
F	34.35	13.57	0.15	0.99			
Р	< 0.01	< 0.01	0.70	0.33			

**Table 4** Prey consumption and oviposition by Amblydromella caudiglans and Galendromus occidentalisfed on two different diets

Numbers in parentheses are the asymmetric 95 % confidence limits

*occidentalis* in commercial apple orchards. Additionally, *A caudiglans*, unlike *G. occidentalis*, has difficulty navigating the webbing produced by *Tetranychus* spp. females and can even become entangled and die in spider mite colonies (Putman 1962; Sabelis and Bakker 1992; McMurtry and Croft 1997). The shift to *P. ulmi* (which produces little webbing) as the most common pest species may have allowed for an increase in *A. caudiglans* populations.

## Pesticide toxicity

Mortality was not significantly different from the check for novaluron, spinetotetramat, and chlorantraniliprole for *A. caudiglans* (Table 5). For *G. occidentalis*, all pesticides except imidacloprid and spinetoram did not cause mortality different from the check (Table 5). Comparing mortality for these species emphasizes that although some pesticides were

Treatment	A. caudiglans n	<i>G. occidentalis</i> % Mortality	% Runoff	n	% Mortality	% Runoff
Carbaryl	25	100a	12.00bc	23	17.39b	13.04b
Azinphosmethyl	24	83.33b	45.83a	18	27.78b	16.67b
Imidacloprid	20	85.00b	20.00b	23	78.26a	65.22a
Spinetoram	22	95.45ab	50.00a	22	90.91a	50.00a
Novaluron	24	25.00 cd	20.83b	24	0.00c	0.00c
Spirotetramat	20	20.00 cd	20.00b	21	9.52bc	9.52bc
Chlorantraniliprole	21	0.00e	0.00c	25	8.00bc	8.00bc
Bifenazate	23	47.83c	43.48a	24	8.33bc	8.33bc
Check	25	8.00de	8.00bc	24	8.33bc	8.33bc
$\chi^2  (df = 8)$		143.03			103.11	
Р		< 0.01			< 0.01	

Table 5 Toxicity of orchard pesticides to Amblydromella caudiglans and Galendromus occidentalis

For pesticide concentrations, see Table 1

acutely toxic to both species (imidacloprid, spinetoram), several pesticides were toxic to *A*. *caudiglans* and not *G. occidentalis* (carbaryl, azinphosmethyl, bifenazate) (Table 5).

OP resistance is well-documented in *G. occidentalis* (Croft and Jeppson 1970; Ahlstrom and Rock 1973; McMurtry 1981); carbaryl resistance has also been reported, but to a lesser extent (Babcock and Tanigoshi 1988; Beers and Schmidt 2014). OP use was documented to cause a replacement of *A. caudiglans* by *G. occidentalis* in an apple orchard in British Columbia (Downing and Moilliet 1967). These studies provide further evidence that the resistance of *G. occidentalis* to older classes of insecticides may have allowed for its previous uniform dominance in Washington commercial apple orchards.

The registration and implementation of mating disruption caused a profound change in the apple pest management program in the last 25 years. During its peak use period, azinphosmethyl was applied to 94 % of Washington's apple acreage, during an era when nine additional OPs and three carbamates were also used (NASS 1991, 1994). The seasonlong coverage of these toxic materials ensured that only an OP-adapted predator could survive. OP used steadily declined during this period, and many of their uses were replaced by more selective materials. Azinphosmethyl was last used in 2013, leaving only three OPs in use, and on greatly decreased acreage (<50 %) (NASS 2011). The dominant replacement codling moth insecticide, chlorantraniliprole (NASS 2011), is the least toxic to both species of predatory mites (Table 5, Beers and Schmidt 2014). The total number of applications for codling moth also declined as mating disruption reduced the pest pressure. While many of the newer pesticides are toxic to predatory mites, the frequency (and in the case of imidacloprid, a lower use rate) provided more opportunities for other species to colonize orchards.

Bifenazate is an interesting case in that it is considered a selective acaricide; the label lists five species of predatory mites (including *G. occidentalis*) which are not adversely affected by the product. However, it did cause significant mortality in *A. caudiglans*. The absence of *A. caudiglans* from the list of species tested is understandable, since its discovery as an important predator in commercial orchards is very recent; however, it emphasizes the importance of species-specific testing for nontarget effects of reduced-risk pesticides. In terms of the population dynamics of these two phytoseiids, bifenazate has been correlated with both higher *G. occidentalis* populations and lower *A. caudiglans* populations (Schmidt-Jeffris et al. 2015). Use of this pesticide clearly has strong potential to drive competition in favor of *G. occidentalis*.

For many of the pesticides tested, the most impactful negative effects on *A. caudiglans* were acute, rather than sublethal; this is in contrast to previous work with *G. occidentalis* (Beers and Schmidt 2014). Sublethal effects (with greatly reduced acute toxicity) of pesticides to *G. occidentalis* are possibly evidence of resistance development. The differences in mortality are likely a mechanism capable of shifting competition in favor of *G. occidentalis* in orchards where these pesticides are sprayed, even if *G. occidentalis* experiences some sublethal effects (e.g., azinphosmethyl) (Beers and Schmidt 2014). The exception would be pesticides that cause overwhelming sublethal effects in *G. occidentalis* (e.g., spirotetramat) (Beers and Schmidt 2014).

Analysis of runoff indicated that azinphosmethyl, spinetoram, and bifenazate were irritant to *A. caudiglans*, whereas imidacloprid and spinetoram were irritant to *G. occidentalis* (Table 5). The repellency assay confirms that all three pesticides causing significant runoff were also repellent to *A. caudiglans* (Table 6). These results further highlight the sensitivity of *A. caudiglans* to bifenazate and azinphosmethyl, whereas both species were sensitive to spinetoram. In contrast, previous research with *G. occidentalis* found little evidence of spinetoram irritancy or repellency (Beers and Schmidt-Jeffris 2015); it

Table 6         Repellency of orchard           pesticides to Amblydromella	Pesticide	Evaluation	n	% on treated half	z*
caudiglans	Azinphosmethyl	1	48	4.17	-3.5
		2	48	10.42	-3.1
		3	34	11.76	-3.2
	Spinetoram	1	45	22.22	-4.3
	-	2	38	23.68	-4.6
		3	27	14.81	-3.8
	Bifenazate	1	45	20.00	-3.7
		2	46	15.22	-4.2
*In all area $P < 0.01$		3	46	4.35	-4.3

In all case, P < 0.01

appears that the effects of this pesticide may vary with population. Imidacloprid is known to be repellent to G. occidentalis (Bostanian et al. 2009) and in our study (where mortality was calculated as dead + runoff), runoff made a larger contribution to total mortality. The opposite was true for A. caudiglans, indicating that this is yet another pesticide where the acute effects seem to be more pronounced for this phytoseiid than G. occidentalis.

# Conclusions

These results provide experimental evidence supporting a recent Washington survey (Schmidt-Jeffris et al. 2015), and previous work with these species in other regions (Downing and Moilliet 1972). While G. occidentalis can thrive under intensive OP use, several of the newer pesticides have the ability to decrease A. caudiglans or G. occidentalis densities. A. caudiglans also cannot survive on a diet of eggs of a Tetranychus spp., and has difficulty navigating their dense webbing. Thus it is possible that a change in both pesticide regimes (away from OPs) and dominant prey species (to P. ulmi) provided the conditions that allowed this predator to become more common in Washington apple orchards in recent years.

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**Conflict of interest** The authors declare that they have no conflict of interest. All procedures performed in studies involving animals were in accordance with the ethical standards of the institution or practice at which the studies were conducted.

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