

# Impact of intraguild predation and lambda-cyhalothrin on predation efficacy of three acarophagous predators

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**Abstract:** This laboratory study reports the interaction of three predators found in commercial apple orchards in Quebec, *Hyaliodes vitripennis* (Say) (Hemiptera: Miridae), *Harmonia axyridis* Pallas (Coleoptera: Coccinellidae) and *Amblyseius fallacis* (Garman) (Acarina: Phytoseiidae). First, intraguild predation between *H vitripennis* and the two other predators was characterized in the absence and presence of their extraguild prey, *Tetranychus urticae* Koch. The results showed an asymmetrical interaction in favour of the larger predator and the levels of intraguild predation were weak for the two predatory combinations. Presence of the phytophagous mite reduced the intensity of intraguild predation in the predatory combination of *H axyridis* and *H vitripennis*. Second, the effects of intraguild predation and the application of lambda-cyhalothrin on predation efficacy of the predators were evaluated. The application of the insecticide reduced prey consumption of *H vitripennis* and *H axyridis* but did not affect that of *A fallacis*. Combination of predators and an insecticide application resulted in two different situations depending on the species involved: a reduced predation efficacy for the combination of *H vitripennis* and *H axyridis* due to a knockdown effect caused by the insecticide, and no effect on *T urticae* consumption for *H vitripennis* and *A fallacis*. It is suggested that an integrated pest management program based on *H vitripennis*, *A fallacis* and lambda-cyhalothrin may be evaluated to repress phytophagous mites in Quebec orchards.

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**Keywords:** intraguild predation; lambda-cyhalothrin; predation efficacy; *Hyaliodes vitripennis*; *Harmonia axyridis*; *Amblyseius fallacis*; *Tetranychus urticae*

## 1 INTRODUCTION

In Quebec commercial apple orchards, pesticides are used to manage arthropod pests and diseases whenever required.<sup>1,2</sup> Among the key pests, phytophagous mites may delay tree growth, reduce fruit size and fruit quality, and induce premature drop of fruit.<sup>3</sup> These damages are linked to the timing of mite injury,<sup>4</sup> the cultivars<sup>5</sup> and the mite population density.<sup>6</sup> Repeated pesticide treatments may decrease in efficacy because of the development of resistance in phytophagous mites and the negative impact of these compounds on the predator populations.<sup>7</sup>

The efficacy of a biological control program for mites may be altered by chemical applications. Pesticide treatments can affect different characteristics

of arthropods, such as fertility, fecundity, longevity, developmental rate, mobility, foraging efficacy and oviposition.<sup>8–13</sup> Pesticides may also modify arthropod behaviour and consequently predator interactions, but few studies have evaluated the impact of pesticides on predation efficacy and on predator interaction. Roger *et al*<sup>12,13</sup> have recorded a reduction of predation efficacy by the ladybeetle *Coleomegilla maculata lengi* Timb in the presence of pesticides due to a reduction of time spent searching for prey. Moreover, prey contaminated by pesticides may be rejected by a predator. Consequently, the efficiency of a biological control agent may be reduced.<sup>14,15</sup>

The efficacy of a biological control program for phytophagous mites may also depend on the

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Contract/grant sponsor: le Fonds pour la Formation de Chercheurs et l'Aide à la recherche du Québec (FCAR)

Contract/grant sponsor: Institut de recherche et de développement en agroenvironnement (IRDA)

(Received 1 June 2004; revised version received 19 October 2004; accepted 3 November 2004)

Published online 15 February 2005

interactions within the guild of natural enemies of these pests. Interaction between predators, such as competition and intraguild predation, may affect the implementation of a new biological agent to manage phytophagous mites. Intraguild predation is an interaction of competition and/or predation among intraguild predators, which share a common prey, resulting in the consumption or death of one of the predators.<sup>16</sup> Hence, intraguild predation may affect the composition, distribution, abundance and evolution of species<sup>16,17</sup> and may be the cause of failure in an integrated pest management program.<sup>18</sup> However, intraguild predation may also have a stabilizing effect on populations of an ecosystem.<sup>19</sup>

*Hyaliodes vitripennis* (Say) (Hemiptera: Miridae) has been reported to be an effective predator of phytophagous mites in Quebec apple orchards.<sup>20,21</sup> It feeds mainly on phytophagous mites and aphids,<sup>22,23</sup> as well as on other arthropods, such as predaceous mites, leafhoppers and Lepidoptera.<sup>22</sup> Other acarophagous predators in apple orchards include *Amblyseius fallacis* (Garman) (Acarina: Phytoseiidae) and *Harmonia axyridis* Pallas (Coleoptera: Coccinellidae).<sup>24</sup> These predators have specific features of interest for integrated pest control programs against phytophagous mites. *Amblyseius fallacis* is a key predator of phytophagous mites, and it has developed resistance to organophosphate and pyrethroid insecticides used in apple orchards.<sup>25–27</sup> *Harmonia axyridis* is a voracious ladybeetle that can consume 40–50 phytophagous mites per day.<sup>28</sup>

In a previous study, we demonstrated that intraguild predation intensity and symmetry between *H vitripennis* and *H axyridis* or *A fallacis* was not affected by a sublethal dose of lambda-cyhalothrin, but that their mobility was modified depending on the predator species.<sup>29</sup> We noted a knockdown effect on *H axyridis* and *H vitripennis* and a higher mobility on *A fallacis*. Another study showed that mortality of the intraguild predator was higher after the application of lambda-cyhalothrin because the predator was also adversely affected by the consumption of contaminated prey.<sup>30</sup>

The present study characterized the intraguild predation between *H vitripennis* and *H axyridis*, and between *H vitripennis* and *A fallacis*, in the absence and presence of extraguild prey. It also examined the effects of intraguild predation and of a sublethal dose of lambda-cyhalothrin on the predation efficacy of the acarophagous guild.

## 2 EXPERIMENTAL

### 2.1 Biological material

*Hyaliodes vitripennis* was collected in a commercial apple orchard at Rougemont (73°03'W, 45°26'N) Quebec, Canada, and maintained in the laboratory 1–2 days on *Tetranychus urticae* Koch. *Harmonia axyridis* was collected from hibernation sites in the same area and reared in the laboratory on *Ephestia kuehniella* Zeller eggs. *Amblyseius fallacis* was obtained

from a permanent colony reared on *T urticae* at the Agriculture and Agri-Food Canada (St-Jean-sur-Richelieu) research facilities. *Tetranychus urticae* was reared on Lima bean leaves. Rearing conditions for all arthropods were 20 ( $\pm$ 1) °C, 60–70% RH and 16:8 h light:dark photoperiod. All tests were carried out in laboratory under similar conditions [24 ( $\pm$ 1) °C, 60–70% RH].

### 2.2 Characterization of intraguild predation

Evaluation of intraguild predation was done in the absence and presence of an extraguild prey, *T urticae*. Experiments were conducted on vertical apple shoots, with three leaves. Shoots were collected from apple trees (cv McIntosh) in the same orchard, washed before the beginning of the experiment in order to eliminate other arthropods and contaminants, and placed in a 4-litre plastic box. Predators were starved for a period of 16 h (photoperiod: 8:8 light:dark) before tests, in order to increase their motivation to forage.

The level of intraguild predation was determined for two predator combinations: (1) *H vitripennis* and *H axyridis*, and (2) *H vitripennis* and *A fallacis*. Combinations of one individual of each species were selected according to two main factors, the developmental stage and predator mobility (sessile and mobile). Non-predatory combinations and combinations where the probability of encounter between the two predators was low were not studied. Eighteen combinations were tested. The combinations included mobile life stages of *H vitripennis* (nymph II, nymph IV and adult) with selected developmental stages of *H axyridis*: egg, larva II, larva IV, pupa and adults, as well as adults of *A fallacis*.

An individual of each predator species of the selected combination was introduced on separated leaves. Combinations involving eggs included a mass of five eggs placed on the ventral side of the leaf. Two treatments were applied: absence of extraguild prey (*T urticae*), and presence of extraguild prey. In the presence of extraguild prey, a Lima bean leaf containing 40 eggs and mobile stages of *T urticae* were placed on the dorsal side of the leaf 16 h before the beginning of the test to allow extraguild prey dispersion on the apple shoot.

After 7 h, predators were removed and mortality and consumption were recorded. Mortality and consumption of eggs and pupae were determined when egg hatch and adult emergence were completed [24 ( $\pm$ 1) °C, 60–70% RH, 16:8 h light:dark]. Natural predator mortality was determined by a control treatment (one predator and no extraguild prey). Fifteen replicates were carried out for each treatment.

The corrected mortality, attributed to predator interaction, was calculated according to Soares *et al*:<sup>31</sup>

$$P = (t - a) * ra$$

where  $P$  = number of replicates with predation (intraguild predation),

$t$  = total number of replicates (=15),  
 $a$  = number of replicates where the individual was alive,  
 $ra$  = ratio of alive individuals in the control.

An index of symmetry (SI: corrected mortality in which a given predator was eaten over the total number of replicates in which there was intraguild predation) was calculated for each pair of predators<sup>32</sup> and it was compared with a theoretical index of 50% (corresponding to a symmetric interaction) using a test of conformity.<sup>33</sup> For each predatory pair, the level of intraguild predation (IL: corrected mortality over the total number of replicates) was calculated.<sup>32</sup> Levels of intraguild predation were cumulated for a specific developmental stage and compared between the different life stages of the selected predator. To determine the vulnerability of the developmental stages, the level of intraguild predation was compared: (1) among developmental stages of *H vitripennis*, and (2) among developmental stages of *H axyridis*, for a specific treatment (the absence or presence of extraguild prey) using a likelihood ratio  $G$  test.<sup>33</sup> In the presence of a significant difference among the predator developmental stages, subsequent  $G$  tests were carried out. The levels of intraguild predation in the absence or the presence of extraguild prey were compared for each developmental stage, using a likelihood ratio  $G$  test.<sup>33</sup> The mean number of eggs consumed by the different life stages of *H vitripennis* (compared with the control treatment) and the impact of the extraguild prey (absence versus presence) on egg consumption were compared with a Wilcoxon–Mann–Whitney test.<sup>33</sup>

### 2.3 Impact of intraguild predation and lambda-cyhalothrin application on predation efficacy

The predation efficacy, corresponding to the number of prey consumed for a fixed period, was evaluated for the three predators separately and in combination to evaluate the impact of intraguild predation. The experiments were conducted on apple leaves collected in an orchard and placed with the dorsal side towards a thin layer of agar in a Petri dish. The agar prevented desiccation of the leaf. Before each test, predators were starved for a period of 16 h.

Twenty adults of *T urticae* were introduced on each leaf. The Petri dish was sealed with a transparent tightened plastic film. Predation efficacy was evaluated for five treatments: (1) one nymph III of *H vitripennis*, (2) one larva III of *H axyridis*, (3) one adult of *A fallacis*, (4) one nymph III of *H vitripennis* with one larva III of *H axyridis* and (5) one nymph III of *H vitripennis* with one adult of *A fallacis*. Predation efficacy was evaluated in the absence and presence of a sublethal dose of lambda-cyhalothrin (Warrior® 120 g litre<sup>-1</sup> EC). Lambda-cyhalothrin is a pyrethroid insecticide commonly used in apple orchards. After the introduction of the predator(s), lambda-cyhalothrin was sprayed on the experimental leaf with a thin-layer chromatography sprayer set at 10.34 kPa at a

distance of 25 cm. The sublethal dose applied was 0.0015 g AI litre<sup>-1</sup>, corresponding to the LC<sub>25</sub> for *H vitripennis*.<sup>34</sup> After 4 h of exposure, predator(s) was (were) removed, predator mortality was noted and *T urticae* were counted. The control treatment consisted only of *T urticae* to evaluate the potential of escape and the natural mortality of the phytophagous mite. Ten replicates were carried out per treatment.

Two theoretical values were calculated by summing the results of monospecific treatments (1: *H vitripennis* + *A fallacis*; 2: *H axyridis* + *H vitripennis*) in order to assess the impact of intraguild predation on predation efficacy. Statistical analyses were carried out with seven treatments. Two-way ANOVA was used (treatment and insecticide application) to evaluate the impact of intraguild predation and insecticide application on predation efficacy.<sup>33</sup> In presence of significant difference, *post hoc* Tukey–Kramer tests were done.

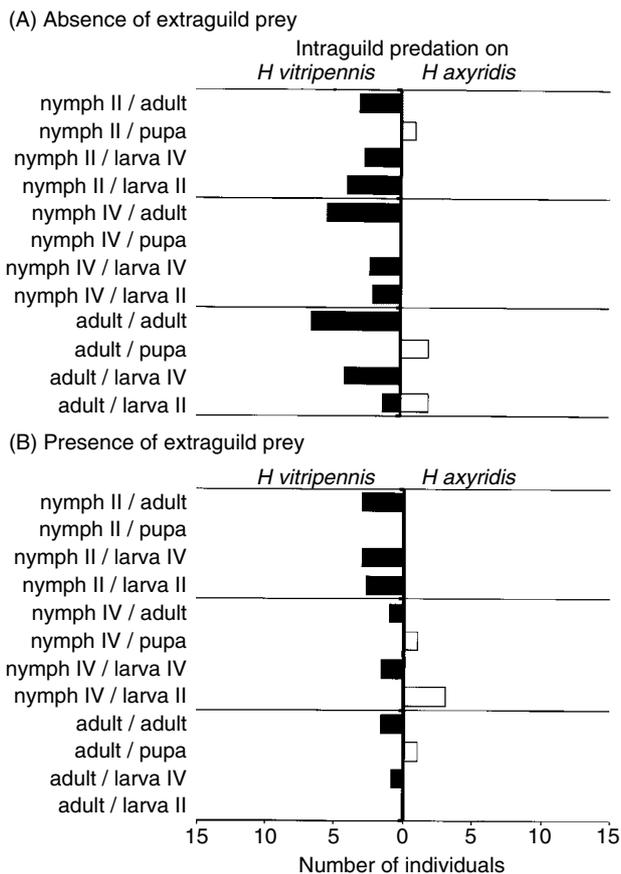
## 3 RESULTS

### 3.1 Characterization of intraguild predation

#### *Hyaliodes vitripennis* versus *H axyridis*

In the absence of *T urticae*, intraguild predation between *H vitripennis* and *H axyridis* was recorded in approximately 20% of the replicates (Fig 1A). Asymmetrical interaction in favour of *H axyridis* was observed in 67% of the combinations (*H vitripennis* nymph II:  $\chi^2 = 8.549$ ,  $df = 1$ ,  $P = 0.0035$ ; *H vitripennis* nymph IV:  $\chi^2 = 30.188$ ,  $df = 1$ ,  $P < 0.0001$ ). We also observed similar levels of intraguild predation on the different developmental stages of *H vitripennis* by *H axyridis* ( $G_2 = 0.240$ ,  $P = 0.8868$ ). *Harmonia axyridis* pupae were attacked more by *H vitripennis* than larvae IV and adults (pupa/larva IV:  $G_1 = 4.262$ ,  $P = 0.0390$ ; pupa/adult:  $G_1 = 4.262$ ,  $P = 0.0390$ ). Eggs of *H axyridis* [larvae emerged in control:  $3.21 \pm (0.27)$ ] were consumed by *H vitripennis* nymphs IV [larvae emerged:  $1.27 (\pm 0.32)$ ] (Wilcoxon,  $Z = 3.61$ ,  $P = 0.0003$ ) and adults [larvae emerged:  $2.27 (\pm 0.37)$ ] (Wilcoxon,  $Z = 1.99$ ,  $P = 0.0456$ ).

In the presence of *T urticae*, predation interaction between *H vitripennis* and *H axyridis* was observed in approximately 10% of the replicates (Fig 1B). Levels of intraguild predation for the three developmental stages of *H vitripennis* by *H axyridis* were similar ( $G_2 = 4.892$ ,  $P = 0.0866$ ). However, we noted higher intraguild predation by *H vitripennis* on *H axyridis* larvae II than on larvae IV and adults (larva II/larva IV:  $G_1 = 4.262$ ,  $P = 0.0390$ ; larva II/adult:  $G_1 = 4.262$ ,  $P = 0.0390$ ). Larval emergence of *H axyridis* in the presence of *H vitripennis* adults [larvae emerged:  $2.13 (\pm 0.42)$ ] was significantly lower than the control [larvae emerged:  $3.21 (\pm 0.27)$ ] (Wilcoxon,  $Z = 2.06$ ,  $P = 0.0393$ ). The presence of the extraguild prey significantly reduced intraguild predation of *H vitripennis* adults ( $G_1 = 5.464$ ,  $P = 0.0194$ ) by *H axyridis*, the level of intraguild predation was reduced by 27% on the mirid nymphs IV ( $G_1 = 3.473$ ,  $P = 0.0624$ ). The predation on *H axyridis* eggs



**Figure 1.** Levels of intraguild predation (IL) of different life stage combinations of *Harmonia axyridis* and *Hyaliodes vitripennis*: (A) in the absence of extraguild prey, *Tetranychus urticae*; (B) in the presence of extraguild prey. Open bars show intraguild predation on *H. vitripennis* and shaded bars show intraguild predation on *H. axyridis*.

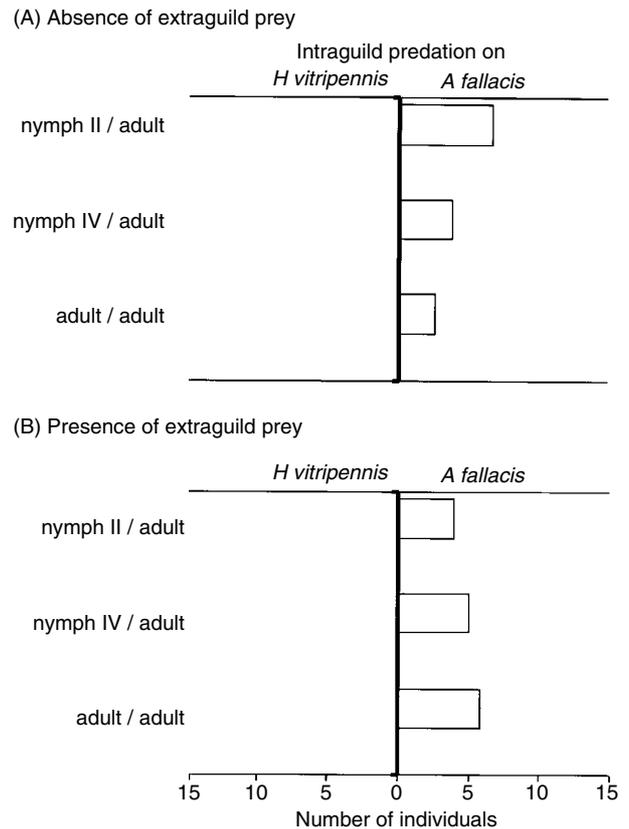
by *H. vitripennis* nymphs IV was also lower than in the absence of *T. urticae* (Wilcoxon,  $Z = 2.25$ ,  $P = 0.0244$ ).

#### *Hyaliodes vitripennis* versus *A. fallacis*

In absence of *T. urticae*, intraguild predation between *H. vitripennis* and *A. fallacis* was present in approximately 30% of replicates and the interaction was always asymmetrical in favour of the mirid (Fig 2A). Intraguild predation levels were similar for the three predatory combinations ( $G_2 = 2.66$ ,  $P = 0.2643$ ). In presence of *T. urticae*, intraguild predation between *H. vitripennis* and *A. fallacis* was observed in approximately 35% of replicates and also asymmetrical in favour of the mirid (Fig 2A). Levels of intraguild predation by the mirid were similar for all combinations ( $G_2 = 0.63$ ,  $P = 0.7397$ ).

### 3.2 Impact of intraguild predation and lambda-cyhalothrin application on predation efficacy

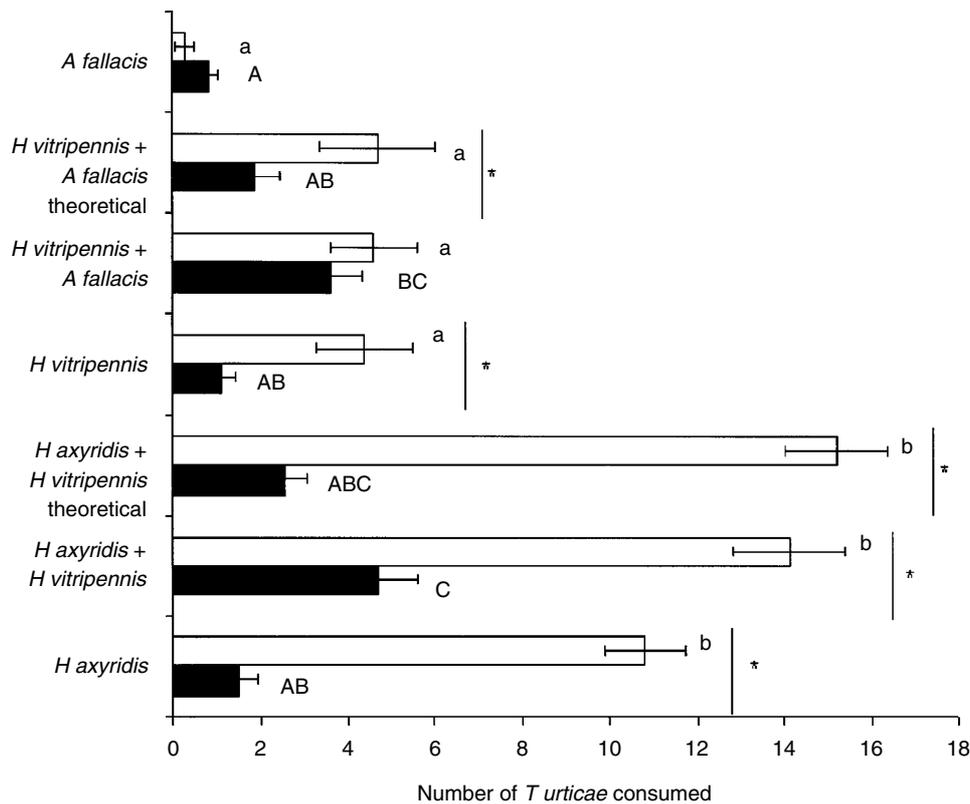
Predation efficacy of the predator combinations were affected by the application of lambda-cyhalothrin (ANOVA 2, model:  $F_{13,136} = 31.72$ ,  $P < 0.0001$ ; predator combination:  $F_6 = 28.98$ ,  $P < 0.0001$ ; insecticide application:  $F_1 = 138.43$ ,  $P <$



**Figure 2.** Levels of intraguild predation (IL) of different life stage combinations of *Hyaliodes vitripennis* and *Amblyseius fallacis*: (A) in the absence of extraguild prey, *Tetranychus urticae*; (B) in the presence of extraguild prey. Open bars show intraguild predation on *A. fallacis*.

0.0001; predator combination  $\times$  insecticide application:  $F_6 = 16.68$ ,  $P < 0.0001$ ). In the absence of lambda-cyhalothrin, *H. axyridis* and *H. axyridis* + *H. vitripennis* were the more efficient treatments to repress *T. urticae* (Tukey–Kramer,  $P < 0.05$ ) (Fig 3). In the presence of the insecticide, *H. axyridis* and *H. axyridis* + *H. vitripennis* were also the more efficient treatments to manage the phytophagous mite (Tukey–Kramer,  $P < 0.05$ ).

Lambda-cyhalothrin did not kill *T. urticae* in the control treatment and all the predators were alive after 4 h. The insecticide affected the three predators differently (Fig 3). *Harmonia axyridis* consumed *T. urticae* seven times more in absence than in presence of lambda-cyhalothrin ( $F_{1,19} = 81.00$ ,  $P < 0.0001$ ). *Hyaliodes vitripennis* consumed 75% more *T. urticae* in the absence than in the presence of lambda-cyhalothrin ( $F_{1,19} = 8.08$ ,  $P = 0.0108$ ). In contrast, predation efficacy of *A. fallacis* was not affected by the lambda-cyhalothrin ( $F_{1,19} = 2.32$ ,  $P = 0.1451$ ). The predation efficacy of the more efficient combination (*H. vitripennis* and *H. axyridis*) was reduced by 67% in presence of lambda-cyhalothrin ( $F_{1,19} = 35.66$ ,  $P < 0.0001$ ), whereas the predation efficacy of *H. vitripennis* and *A. fallacis* was not adversely affected ( $F_{1,19} = 0.62$ ,  $P = 0.4407$ ).



**Figure 3.** Predation efficacy of different predatory combinations, in the absence (open bars) and the presence (shaded bars) of lambda-cyhalothrin. Lower case letters indicate a significant difference between predatory treatments in absence of lambda-cyhalothrin (ANOVA,  $P < 0.05$ ). Upper case letters indicate a significant difference between predatory treatments in presence of lambda-cyhalothrin (ANOVA,  $P < 0.05$ ). Asterisks indicate a significant difference of predation efficacy in absence versus in presence of insecticide application (ANOVA,  $P < 0.05$ ).

#### 4 DISCUSSION

Our experiments have produced two main conclusions. First, we observed intraguild predation between *H vitripennis* and *H axyridis*, as well as between *H vitripennis* and *A fallacis*. However, the intensity of the interaction was weak. Second, following the application of lambda-cyhalothrin, the *H vitripennis* and *A fallacis* combination consumed more *T urticae* than each predator alone, while no such effect was observed in the *H axyridis* and *H vitripennis* combination. Thus, the application of lambda-cyhalothrin affected the predators and their interactions differently.

Intraguild predation for the two predatory combinations was generally asymmetrical in favour of the larger predator. In the combination of *H axyridis* and *H vitripennis*, we noted intraguild predation in less than 20% of the replicates, and in the combination of *H vitripennis* and *A fallacis*, intraguild predation was observed in approximately 30% of the replicates. Lucas et al<sup>32</sup> and Agarwala and Dixon<sup>35</sup> showed higher levels of intraguild predation in Coccinellidae, between 50 and 100%. Moreover, the intensity of intraguild predation in the combination of *H axyridis* and *H vitripennis* was reduced in the presence of *T urticae*. Lucas et al,<sup>32</sup> Yasuda and Shinya<sup>36</sup> and Agarwala and Dixon<sup>35</sup> have demonstrated a reduction of the intraguild predation intensity between two predators when they were placed in the presence of an extraguild

prey. The direction of our intraguild predation interactions was determined by size and vulnerable stages. The larger predator usually attacked the smaller one. The predator size ratio had been shown to influence intraguild predation in Coccinellidae, Heteroptera and Acari.<sup>32,37-40</sup> Young stages and immobile stages were susceptible to intraguild predation. Eggs and pupa were vulnerable sessile stages as they did not have active defence mechanisms. This resulted in higher susceptibility to predation.<sup>35,36,41</sup> Intraguild predation has frequently been observed on immature and immobile stages because their defensive mechanisms are less effective and their mobility reduced.<sup>42</sup>

Our results indicated that in the absence of lambda-cyhalothrin, the intensity of intraguild predation between *H axyridis* and *H vitripennis* and between *H vitripennis* and *A fallacis* was low, and the interaction did not affect the predation efficacy. Thus an additive effect was reported on the shared prey. Different types of response have been reported from the literature: (1) the pest was not controlled efficiently when several predators had been used because of intraguild predation and/or competition,<sup>43,44</sup> (2) an additive or synergistic (more than additive) effect was observed on the shared prey when several generalist predators were used together<sup>45-48</sup> and (3) different results were obtained depending on the life stages<sup>49</sup> or predatory species<sup>39,50</sup> involved.

The impact of lambda-cyhalothrin application differed depending on the treatment. In the predatory combination of *H axyridis* and *H vitripennis*, the number of *T urticae* consumed was three times lower than in the absence of the insecticide treatment, whereas, in the *H vitripennis* and *A fallacis* combination, consumption of *T urticae* was not affected by the application of lambda-cyhalothrin. Such results may be due to the impact of the insecticide on: (1) the extraguild prey (*T urticae*), (2) the intraguild predator, (3) the intraguild prey, (4) the intraguild predation level and (5) on a combination of previous effects. No mortality of *T urticae* was caused by lambda-cyhalothrin application. The intraguild predators and intraguild preys were affected differently by the insecticide application; we noted a reduced predation efficacy of *H axyridis* and *H vitripennis*, whereas consumption of *T urticae* by *A fallacis* was not affected by the insecticide application. A previous study demonstrated that the mobility of *H axyridis* and *H vitripennis* was reduced after exposure to lambda-cyhalothrin while *A fallacis* increased its time spent moving post-application.<sup>29</sup> Moreover, we observed a knockdown effect of lambda-cyhalothrin on *H axyridis*, the coccinellid being inactive for a long period following the insecticide application. The reduced mobility and the knockdown effect can explain the lower consumption of *T urticae* by *H axyridis* and *H vitripennis* in the presence of this insecticide. Increased movements of *A fallacis* may suggest an increased predation efficacy; however, the higher mobility could also be explained as an escape behaviour<sup>11</sup> caused by the repellent properties of lambda-cyhalothrin.<sup>51</sup>

In our predation efficacy experiments, no intraguild predation was noted during the test period. Predation efficacy of *H axyridis* and *H vitripennis* in combination was significantly reduced after the insecticide application. The predation efficacy of each predator was reduced by the application of lambda-cyhalothrin and resulted in lower suppression of *T urticae*. Predation efficacy of the *H vitripennis* and *A fallacis* combination was not affected by the insecticide application. Few studies have evaluated the combination of predators and pesticides on pest suppression. Fagan *et al*<sup>47</sup> have reported effective pest control when the treatment (predator or pesticide) was applied alone, but when applied in concert, these treatments cancelled the effect of each other. These authors suggested that the failure of the control of the target pest was caused by the elimination of a secondary predator by the pesticide and intraguild predation. In an earlier study, the application of lambda-cyhalothrin did not significantly affect the intensity of intraguild predation between *H axyridis* and *H vitripennis* and between *H vitripennis* and *A fallacis*.<sup>29</sup> However, in the present study, the consumption of *T urticae* by the combination of *H vitripennis* and *A fallacis*, was slightly increased after insecticide application (3.8) when compared with the individual consumption of each predator alone (summed, 1.9). It is likely that the lambda-cyhalothrin

did not affect the intensity of intraguild predation but influenced the consumption of *T urticae* when the two predators were combined, generating a more than additive effect on the shared prey.

## ACKNOWLEDGEMENTS

We thank F Courchesne for technical assistance. This research was supported by a grant from le Fonds pour la Formation de Chercheurs et l'Aide à la recherche du Québec (FCAR) to D Coderre, G Chouinard and NJ Bostanian. C Provost was supported by graduate scholarships from Institut de recherche et de développement en agroenvironnement (IRDA). We are grateful to Agriculture and Agri-food Canada for providing the predacious mites and to Syngenta Crop Protection Canada Inc for graciously providing lambda-cyhalothrin.

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