CONTROL OF THE MULTICOLOURED ASIAN LADY BEETLE, *HARMONIA AXYRIDIS* (PALLAS) (COLEOPTERA:COCCINELLIDAE) ON GRAPES IN ONTARIO

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Abstract *J. ent. Soc. Ont.* 135: 119–123

The multicoloured Asian lady beetle, *Harmonia axyridis* (Pallas) (Coleoptera: Coccinellidae), has become a pest of grapes, *Vitis vinifera L.* (Rhamnales: Vitaceae), in Ontario because beetles present in harvested grapes produce alkaloids (methoxypyrazines) when stressed or as a pheromone, and these can affect the quality of wines and juices. We have assessed the use of selected insecticides in both laboratory and field to determine the potential for control of this pest. Although, the neonicotinoid acetamiprid was most toxic in the laboratory, malathion or cypermethrin were most effective in the field. Malathion was less persistent than cypermethrin; effects were lost within 7 days whereas cypermethrin residues reduced beetle numbers in plots for at least 7 days.

Introduction

The multicoloured Asian lady beetle , *Harmonia axyridis* (Pallas) (Coleoptera: Coccinellidae), is common in southern Ontario, and like many other species of lady beetle is normally classed as a valuable beneficial species. Introduced initially into North America from Asia in 1916 and in repeated releases since, it has provided effective biological controls in a number of agricultural ecosystems (pecans, strawberries and roses), and is the most common predator of the Chinese soybean aphid, *Aphis glycines* Matsumura (Homoptera: Aphididae) (cited in Koch 2003).

However, *H. axyridis* has become a pest of fruit crops and grapes in Ontario, because in autumn, beetles aggregate and feed on these crops. Whether the beetles cause the primary damage or feed at sites damaged by other insects or birds is not clear but a recent report (Koch *et al.* 2004) indicates a preference for damaged fruits. However, they have become a serious problem on grapes, *Vitis vinifera L.* (Rhamnales: Vitaceae), because beetles are present in harvested bunches and are crushed with the grapes. *H. axyridis*, and some other species of lady beetle produce methoxypyrazines as both an attractant (pheromone) and as a defense mechanism to protect beetles from predators (Hodek 1973). The quality of wines produced from these infested grapes is seriously affected (Pickering *et al.* 2004) and up to 20% of the wines produced in Ontario in 2001 were tainted (G.M.W, personal communication).

This has created a requirement in some seasons for control of these beetles on grapes grown for both wine and juice in Ontario. As part of an initial response to this problem we have investigated the use of selected insecticides. Most of the literature on the effects of pesticides on *H. axyridis* relates to the impact of insecticides on these insects as a non-target species but there are a number of studies summarized by Koch (2003). Generally, these studies indicated that adults were less sensitive than larval stages. Further, as a group, pyrethroids were most toxic but there were large differences between the various pyrethroids. Carbaryl was rated as highly toxic to adults but few data on the effects of organophosphorus insecticides were presented in Koch's (2003) review. The neonicotinoid acetamiprid was rated highly toxic to eggs, larvae and adults in laboratory studies by Youn *et al.* (2003). Another neonicotinoid, imidacloprid, was less toxic to late instar larvae and pupae.

We have assessed a number of insecticides in laboratory and field but because *H. axyridis* becomes a pest of grape within a few days of harvest, we restricted compounds tested in the field to malathion, acetamiprid and cypermethrin which are registered for use close to harvest (three-seven days preharvest). Carbaryl is allowed up to five days of harvest in Ontario, but residues detected in wine from such uses preclude its use here.

Methods

Laboratory Bioassays

For assays, we used beetles collected from unsprayed vineyards of cv.Riesling in the Vineland area in October 2003. Beetles were collected in polyethylene vials lined with plant leaves, and were held overnight in a refrigerator at 4°C. Insecticides, (malathion, purity 99%, cypermethrin, 98%, permethrin, 98%, and carbaryl 98%) were analytical grade samples (Chem Services, West Chester PA) and acetamiprid, 99.9% a.i., (Bayer Inc, Etobicoke, ON) were applied to beetles in five mL of analytical grade acetone using a Potter spray tower (12 s spray time followed by five s settling time). Controls were treated with five mL acetone. Beetles were anaesthetized with CO2, 30 s at 30 kPa and placed upright in glass Petri dishes on a Whatman #1 filter paper, five/dish, for treatment. Treated beetles were held in 200 mL glass jars for 24h in a rearing room at 22 ± 2°C, 60%RH with a photoperiod of 16:8 (L:D). Beetles unable to walk after prodding with a fine brush were considered dead. For calculation of concentration/ response regressions, six concentrations with ten replicates of five beetles were used. Tests were over several days. Data were subjected to probit analysis (POLO-PC LeOra Software, Berkeley, California). Differences in responses of beetles to the various insecticides were considered different if the 95% confidence limits at the LC50 did not overlap.

To determine whether beetles recovered from treatments of acetamiprid, cypermethrin or malathion, we treated ten replications of five beetles at one of the concentrations used in the calculation of the concentration/response regressions described above and assessed mortality at 24, 48 and 72h post treatment to determine whether mortality changed over time (i.e., whether beetles recovered from treatment or more were affected). Differences in means for the three post treatment times were tested with an analysis of variance.

Field Tests

Tests were conducted in a ca. ten -year-old vineyard of cv. Riesling near Vineland, Ontario, where earlier observations had indicated high numbers of beetles. Plots were located on a south facing edge of the vineyard and clearly held a population of *H.axyridis*. Treatments were replicated four times with plots (five vines/plot) arranged in a randomized complete block design. Insecticides, malathion (Malathion 500EC, United Agri Products, London, ON), cypermethrin, (Cymbush 250EC, Zeneca Agro,Stoney Creek ,ON) and acetamiprid (Assail 70WP, DuPont Canada Inc., Mississauga, ON) were applied 23 October 2003. These insecticides were diluted to a rate comparable to 3000L/ha and sprayed to runoff using a Rittenhouse truck-mounted sprayer (Rittenhouse Sprayers, St Catharines, ON) equipped with a Spraying Systems handgun fitted with a D-6 orifice plate. Pressure was set at 2000 kPa. Each plot required eight to nine L of spray mix. Plots were sampled one day (24 October) and seven days (30 October, 2003) after treatment when total numbers of beetles in bunches and on leaves were recorded for each plot. Data were transformed (log (x+1)), analyzed by ANOVA, and means separated with a Tukey test (P=0.05) .

Results and Discussion

Laboratory bioassays showed that acetamiprid and cypermethrin were most toxic to beetles (Table I). Another pyrethroid, permethrin was less toxic than cypermethrin but was ca. equitoxic to malathion or carbaryl. Observations reported were 24h post treatment. Beetles treated with acetamiprid were affected (appeared immobilized) at concentrations lower than we used in bioassays but these responded as capable of normal movement when prodded. Observations 24, 48 and 72 hours after treatment indicated increased mortality of beetles treated with acetamiprid (Table II). Earlier reports (Williams and Fickle 2003) with another neonicotinoid, imidacloprid, indicated initial knockdown followed by recovery, but this did not occur with acetamiprid. Mortality associated with treatments of cypermethrin or malathion was unchanged in these later observations.

TABLE I. Toxicity of insecticides to field-collected multicoloured Asian lady beetles in the laboratory.

TREATMENT	SLOPE ± SE	LC50 (95% CL)[a] mg/L	χ^2
Acetamiprid[b]	2.4 ± 0.2	22.2 (17.0 - 27.2)	6.4
Cypermethrin	1.4 ± 0.2	28.1 (14.8 - 41.9)	9.8
Permethrin	2.9 ± 0.2	131.5 (108.4 - 154.6)	5.7
Malathion	4.0 ± 0.3	211.1 (129.0 - 287.3)	58.0
Carbaryl	3.6 ± 0.3	91.5 (76.1 - 106.7)	7.2

[a] Mortality 24h after treatment
[b] n = 350

In the field, one day after treatment, plots treated with cypermethrin and malathion had significantly (P=0.05) fewer beetles than untreated plots or those treated with acetamiprid (Table III). A number of beetles on acetamiprid-treated vines were affected similarly to those in the laboratory but crawled readily when placed on the hands of the observers. In other treated plots most dead beetles were on the ground under vines but a few dead individuals were caught up in the grape bunches. In our survey of the literature, we did not find information on whether or not dead beetles are a potential source of methoxypyrazines, or how long these compounds might persist in dead insects. In samples seven days post treatment, plots treated with cypermethrin had fewer beetles than control plots but there were no differences between insecticide treatments (Table III). Whether the numbers surviving in this plot (mean of seven beetles over five vines) are above a threshold which could result in off flavors in wine produced

TABLE II. Influence of holding time on the toxicity of insecticides to multicoloured Asian lady beetles.

TREATMENT	PERCENT MORTALITY Hours after Treatment[a]		
	24h	48h	72h
Acetamiprid 30 ppm	56b[a]	84[b]	76[b]
Cypermethrin 50	70[a]	82[a]	78[a]
Malathion 200	30[a]	32[a]	34[a]

[a] Beetles held at 24°C, 60% RH
[b] Means of 10 replications of 5 beetles: means separated by ANOVA, numbers in same row followed by same letter NSD

TABLE III. Control of multicoloured Asian lady beetles on Riesling grape - 2003.

TREATMENT[a]	RATE g a.i./ha	BEETLES/PLOT DAYS AFTER TREATMENT	
		1	7
Cypermethrin (Cymbush 250EC)	60	0.0[bb]	7.0[b]
Malathion (Malathion 500E)	900	0.3[b]	11.0[ab]
Acetamiprid (Assail 70WP)	56	11.8[a]	27.0[ab]
Control	-	25.8[a]	38.8[a]

[a] Applied 23 October 2003, plots 7.5 long in 2.1 m spaced rows, replicated 4 times.
[b] Numbers in same column followed by same letter are not significantly different $P<0.05$, Tukey test

from these grapes is not known and may require considerable additional study. In any case, it would be advantageous for dead beetles to fall to the ground so they would not be harvested with the grapes.

The lack of control with acetamiprid was surprising given the results of the laboratory assays: the rate we tested in the field was based on rates recommended (Anonymous 2003) on grape for leafhopper control. Reissig (2003) reported that another neonicotinoid, imidacloprid, which was highly toxic in laboratory tests against apple maggot, (*Rhagoletis pomonella* (Walsh)) (Diptera:Tephritidae), was less effective than expected when applied to foliage in the field. Hu *et al.* (1998) speculated that the ineffectiveness of imidacloprid against apple maggot in the field might be associated with rapid absorption and consequent inactivity against insects on the surface or to rapid degradation by sunlight.

Cypermethrin-treated plots had fewer beetles (than controls) in samples both one day and seven days post treatment indicating that toxic residues persisted at least seven days. Malathion reduced beetle numbers in samples one day post treatment, but was not effective in the sample seven days post treatment. Currently, cypermethrin has a seven-day preharvest limit on grape (Anonymous 2003) and appears to be the product of choice based on these trials. However, it is not registered for use in the U.S. and would not be useful on grapes grown for juice for export. Malathion was effective in samples one-day post treatment and can be used up to three days preharvest and might be used on grapes grown for export.

This study has identified two insecticides that reduced infestations of *H. axyridis* on grapes, and which may be used close to harvest when beetles become a problem. This may provide an immediate or short-term response to a new problem but in the longer term, management of beetle populations below a still to be determined threshold by other means is desirable. *H. axyridis* is beneficial throughout much of its life cycle, and effective populations should be preserved.

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