Toxicity of viticultural pesticides to the predatory mites *Amblyseius victoriensis* and *Typhlodromus doreenae*

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Summary

Thirty pesticides used in Australian viticulture were bioassayed in the laboratory for toxicity to the predatory mites *Typhlodromus doreenae* Schicha and *Amblyseius victoriensis* (Womersley). None of 14 synthetic fungicides were toxic to *T. doreenae* but most caused some mortality in *A. victoriensis*. Benomyl, iprodione, chlorothalonil and procymidone were least toxic and mancozeb and triadimefon most toxic, to *A. victoriensis*. The inorganic fungicides, wettable sulphur and copper hydroxide, were non-toxic to *T. doreenae* even at 200 and 33 times field rates, respectively. Copper hydroxide was also non-toxic to *A. victoriensis* but lime sulphur was highly toxic to both species. Insecticides and miticides were generally toxic to both species, particularly *A. victoriensis*. However, azinphos-methyl had low toxicity against *T. doreenae*. Implications of these results for development of biological mite management programs in Australian viticulture using *T. doreenae* and *A. victoriensis*, are discussed.

Introduction

Amblyseius victoriensis (Womersley) and *Typhlodromus doreenae* Schicha (Acari: Phytoseiidae) are responsible for biological control of grapevine mites (*Colomerus vitis* [Pagenstecher], *Calepitrimerus vitis* [Nalepa], *Brevipalpus* spp.) in the Riverland of South Australia (James and Whitney 1993a). *T. doreenae* also regulates grapevine mites in a number of other viticultural regions including the Canberra district, Swan Hill, Mudgee and Cowra (James 1993a, James *et al.* 1995). Mite management programs involving these predators are currently being developed for viticultural regions throughout Australia (James and Whitney 1994). It is essential that their compatibility with pesticides (insecticides/miticides/fungicides) used in grape growing is assessed, so that recommendations can be made regarding the suitability and impact of specific sprays on biological control.

James (1989) examined the impact of eighteen pesticides used in peach cultivation on *A. victoriensis* populations and found six fungicides, three miticides and two insecticides permitted at least some survival of the predator. However, only the miticide clofentezine and fungicides chlorothalonil and diathianon, were considered to have no deleterious effect on survival of *A. victoriensis*. Little is known regarding the susceptibility of *T. doreenae* to pesticides although populations occur in vineyards which use only wettable sulphur and copper for disease control but are usually absent in vineyards exposed to synthetic pesticides (James and Whitney 1993a).

Table 1. Details of pesticides used in toxicity tests against adult female *Typhlodromus doreenae* and *Amblyseius victoriensis*.

Pesticide	Trade Name	Chemical Name	Formulation	Main target pest/disease on grapevines
Fungicides	Benlate®	benomyl	50% wp	Powdery mildew
	Spin®	carbendazim	50% wp	Bunch rot
	Bravo®	chlorothalonil	50% ec	Downy mildew
	Dithane®	mancozeb	80% wp	Downy mildew
	Delane®	dithianon	75% wp	Downy mildew
	Rubigan®	fenarimol	12% ec	Powdery mildew
	Rovral®	iprodione	50% wp	Bunch rot
	Ridomil plus®	metalaxyl + copper oxychloride	15% wp	Downy mildew
	Sumisclex®	procymidone	27.5% ec	Bunch rot
	Tilt®	propiconazole	25% ec	Powdery mildew
	Thiram®	Bis-disulphide	80% wp	Black spot
	Bayfidan®	triadimenol	25% ec	Powdery mildew
	Ronilan®	vinclozolin	50% ec	Bunch rot
	Curit®	Zineb	80% wp	Downy mildew
	Ziram®	Ziram	80% wp	Black spot
	Lansul®	wettable sulphur	80% wp	Powdery mildew
	Lime sulphur®	lime sulphur	21.5% ec	Powdery mildew
	Kocide®	copper hydroxide	50% wp	Downy mildew
	Fungi-fos®	phosphorus acid	20% ec	Downy mildew
	Synertrol oil®	vegetable oil	–	Powdery mildew
Insecticides	Gusathion®	azinphos-ethyl	40% ec	lightbrown apple moth
	Carbaryl®	carbaryl	80% wp	lightbrown apple moth
	Lorsban®	chlorpyrifos	25% wp	lightbrown apple moth
	Diazinon®	diazinon	80% ec	borers, weevils
	Rogor®	dimethoate	40% ec	mites
	Thiodan®	endosulfan	35% ec	thrips
	Maldison®	malathion	50% ec	borers, weevils
	Supracide®	methidathion	40% ec	borers, weevils
Miticides	Carbamult®	promecarb	49% wp	mites
	Kelthane®	dicofol	7.5% wp	mites

wp = wettable powder
ec = emulsifiable concentrate

This paper presents data on the toxicity of a range of pesticides used in Australian viticulture, to *A. victoriensis* and *T. doreenae*.

Materials and methods

Typhlodromus doreenae was cultured on plastic arenas as described by James and Whitney (1993b), from a strain which originated from vineyards in the Renmark/Loxton district of South Australia. Cultures were occasionally boosted by introduction of additional field-collected material and held at 25°C under a 15 hour photophase. Testing was conducted during July 1991 to November 1992. *A. victoriensis* was reared on dwarf bean plants raised in a shadehouse and dusted with the pollen of cumbungi (*Typha orientalis* Presl.) (James 1993b). The strain originated from garden ornamentals in the Leeton/Yanco district (southern New South Wales). The culture was maintained under natural temperatures and photoperiod during October 1992 to August 1993 when testing was conducted. During late autumn–winter, predators were brought indoors and held on arenas for a few days at 20–22°C prior to testing.

Twenty fungicides, eight insecticides and two miticides (Table 1) used in Australian viticulture, were tested in the laboratory for toxicity against adult female *T. doreenae* and *A. victoriensis*. Most products were tested against both species at field and 10 times field rate concentrations (% a.i.), although phosphorus acid, chlorothalonil and synertrol oil were not tested against *T. doreenae* and vinclozolin, zineb and diazinon were not tested against *A. victoriensis*. The inorganic fungicides were tested at concentrations ranging from 0.25–2 times (lime sulphur), 16–33 times (copper hydroxide) and 100–200 times (wettable sulphur) the field rates.

Direct toxicity tests

Predators were removed from rearing arenas or bean leaves and placed on bean leaf discs (38 mm diameter) laid on saturated cotton wool in plastic or metal trays. Manipulation of the predators was conducted using a moistened fine camel hair brush. Ten to fifteen predators were placed on each disc and two or four discs were prepared for each concentration of pesticide tested. A further two to four discs were used as controls. Discs were then sprayed with aqueous suspensions of the pesticide being tested using a Potter Precision Spray Tower. The spraying pressure was 50 kPa and 2 mL of liquid was used for each concentration giving a deposit of 1.6 mg of liquid per square centimetre. Each test was replicated three times (unless otherwise stated) and data for each concentration were derived from a minimum of 45 individuals. Controls, sprayed with water only, were included in each replicate. After spraying, the discs were placed back onto the saturated cotton wool. Discs were supplied with food (cumbungi pollen) and shelter (small piece of broken glass cover slip) for the predators and held at 25±1°C, 60–70% rh, under a 15 hour photophase. Mortality was assessed after 48 hours by gently prodding each predator with a fine bristle. Predators were considered dead if unable to show coordinated movement. Individuals which became entangled in the wet cotton wool were excluded from the analysis. All data were corrected for control mortality (Abbott 1925) and tests discarded if this mortality exceeded 10%.

Table 2. Percentage mortality[A] of adult female *T. doreenae* and *A. victoriensis* sprayed with field rate (FR) and 10 times the field rate concentrations of synthetic fungicides, insecticides and miticides, using a Potter Precision Spray Tower.

Pesticide (FR % a.i.)	*T. doreenae*		*A. victoriensis*	
	FR	10 × FR	FR	10 × FR
Fungicides				
Benomyl (0.02)	1 a	4 a	0	0
Carbendazim (0.05)	0	0	12 a	18 a
Chlorothalonil (0.15)	–	–	7 a	9 b
Dithianon (0.075)	0	4 a	12 a	52 c
Fenarimo (0.0024)	2 a	4 a	19 b	12 b
Iplodione (0.025)	2 a	4 a	0	0
Mancozeb (0.16)	0	0	68 c	100
Metalaxyl (0.02)	4 a	11 b	21 b	75 d
Procymidone (0.05)	0	0	0	4 e
Propiconazole (0.0025)	4 a	5 a	14 a	49 c
Thiram (0.12)	0	17 b	*21 b	*10 b
Triadimenol (0.0025)	2 a	1 a	55 c	84 d
Vinclozolin (0.05)	2 a	0	–	–
Zineb (0.012)	0	3 a	–	–
Ziram (0.012)	5 a	3 a	–	–
Insecticides				
Azinphos-methyl (0.05)	*22 a	*24 a	100	100
Carbaryl (0.10)	36 b	54 b	100	100
Chlorpyrifos (0.025)	100	100	100	100
Diazinon (0.05)	*96 c	100	–	–
Dimethoate (0.05)	100	100	100	100
Endosulfan (0.07)	40 b	97 c	77	100
Malathion (0.10)	15 a	100	100	100
Methidathion (0.05)	100	100	100	100
Miticides				
Carbamult (0.049)	97 a	100	100	100
Dicofol (0.05)	41 b	96	100	100

* Two replicates only, all others are means of three replicates.

[A] Values within pesticide group columns followed by same letter are not significantly different ($P<0.05$).

Residual toxicity tests

The effect of dried deposits of five insecticides and one miticide on mortality of adult female *T. doreenae* and *A. victoriensis* was examined. Leaf discs (28 mm diameter) were cut from grapevine leaves and dipped for five seconds in aqueous suspensions of the pesticide being tested. The discs were then placed on saturated cotton wool and air-dried at 30°C for two hours. Control discs were dipped in water. Ten to 15 predators were introduced onto each disc. Holding conditions and mortality assessment were as described previously. Mortality data were subjected to analysis of variance and least significant difference procedures.

Results

Direct toxicity tests

Typhlodromus doreenae. None of 14 synthetic fungicides used at field rate concentrations were toxic (Table 2). Only two of these compounds, metalaxyl and thiram, showed some toxicity at 10 times field rates. Similarly, the inorganic fungicides, wettable sulphur and copper hydroxide were non-toxic at field rate concentrations (Table 3). Less than 20% mortality occurred when wettable sulphur was used at 100 times the field rate and less than 50% at 200 times the field rate. At these concentrations, predators were thickly coated with sulphur deposits. Copper hydroxide produced no mortality at field

Table 3 Percentage mortality [a] of adult female *T. doreenae* and *A. victoriensis* sprayed with inorganic fungicides using a Potter Precision Spray Tower.

Fungicide and rate %a.i.	*T. doreenae*	*A. victoriensis*
Wettable sulphur		
64.0	46 a	–
32.0	18 b	–
0.64	0	100
0.32*	0	96
Lime sulphur		
4.3	100	–
2.15*	100	100
1.075*	60 a	100
0.54	65 a	–
Copper hydroxide		
10.0	5	–
5.0	0	6
0.3*	0	6
Phosphorus acid		
1.20	–	0
0.12*	–	0
Vegetable oil		
5.0	–	70 a
0.5*	–	6 b

* Registered field rates on grapevine.

[a] Values within pesticide group columns followed by same letter are not significantly different ($P<0.05$).

Table 4. Percentage mortalitya of adult female *T. doreenae* and *A. victoriensis* exposed to field rate (FR) and 10 × field rate residues of insecticides and a miticide in leaf disc bioassays.

Insecticide / miticide (FR % a.i.)	*T. doreenae* FR	*T. doreenae* 10 × FR	*A. victoriensis* FR	*A. victoriensis* 10 × FR
Carbamult (0.049)	100*	100*	100*	100*
Carbaryl (0.10)	56 a	80 a	100	100
Chlorpyrifos (0.025)	100	100	100*	100*
Endosulfan (0.07)	10 b	27 b	80	100
Malathion (0.10)	46 a	88 a	100	100
Methidathion (0.05)	–	–	100*	100*

* Two replicates only.

[a] Values within columns followed by same letter are not significantly different ($P<0.05$).

or 16 times the field rate and only 5% mortality at 33 times the field rate. In contrast, lime sulphur caused substantial mortality at the two recommended field rates. Even at the lowest concentration tested (25% of the upper field rate), 65% of predators were killed.

Most of the insecticides / miticides at field and 10 times field rate concentrations were toxic to *T. doreenae* but three (malathion, azinphos-methyl, carbaryl) allowed reasonable levels of survival (Table 2). Five compounds (methidathion, dimethoate, diazinon, chlorpyrifos and carbamult) produced 96–100% mortality at field rates. Dicofol and endosulfan killed 40–41% of predators at field rates and 96–97% at the higher rates. Malathion permitted good survival at the field rate but killed all predators at the high rate. Carbaryl produced 36% mortality at the field rate and nearly 50% of the predators survived at 10 times the field rate. Azinphosmethyl was least toxic with only 24% mortality at the high rate.

Residual toxicities of insecticides / miticides to *T. doreenae* were similar to those obtained in direct toxicity tests (Table 4), with only carbamult and chlorpyrifos showing high toxicity at field rates.

Amblyseius victoriensis. Four of the synthetic fungicides (benomyl, chlorothalonil, iprodione, procymidone) produced less than 10% mortality at field and 10 times field rates (Table 2). In contrast, mancozeb and triadimenol killed 55–68% of predators at field rates and 84–100% at the higher rates. The remaining compounds permitted 79–88% survival at field rates with carbendazim, fenarimol and thiram also having relatively low toxicity at 10 times field rates.

Wettable sulphur and lime sulphur at field rates were highly toxic to *A. victoriensis* (Table 3). Copper hydroxide and phosphorus acid were virtually non-toxic as was vegetable oil at the field rate.

All insecticides / miticides except endosulfan, caused 100% mortality of *A. victoriensis* at field rates. Twenty three percent of predators survived the field rate of endosulfan but all died at the higher rate. Similar results were obtained in residual tests (Table 4).

Discussion

The data presented here show the toxicity of a range of pesticides used in Australian viticulture, to *T. doreenae* and *A. victoriensis*. These predators provide biological control of mites in South Australian and Canberra district vineyards (James 1993a, James and Whitney 1993a, James *et al.* 1995) and form the basis of mite management programs being developed for all Australian viticultural regions (James and Whitney 1994). Identification of compatibilities between *T. doreenae, A. victoriensis* and viticultural pesticides is a key component of this research.

Typhlodromus doreenae and *A. victoriensis* differed considerably in their susceptibility to the pesticides used in this study. In general, *T. doreenae* had greater tolerance to pesticides, particularly fungicides. All synthetic fungicides tested had low toxicity to *T. doreenae* as did the inorganic fungicides, wettable sulphur and copper hydroxide. The only fungicide to cause high mortality was lime sulphur which is often used early in the growing season. *T. doreenae* overwinters on grapevines (James and Whitney 1993a) and a spring application of lime sulphur is likely to be highly detrimental to the predator. The high tolerance of *T. doreenae* to wettable sulphur is an important attribute. The use of wettable sulphur to control powdery mildew is widespread and increasing in popularity as inputs of synthetic pesticides are reduced. Sulphur resistance was reported in vineyard strains of the phytoseiid *T. occidentalis* Nesbitt in California (Hoy and Standow 1982) but *T. pyri* Scheuten, an important predator in European vineyards, suffered 90% mortality when exposed to 0.25% wettable sulphur in laboratory bioassays (Van de Vrie 1962). It is not known whether sulphur tolerance is inherent in *T. doreenae* or whether it is an example of developed resistance. The Riverland of South Australia where the *T. doreenae* strain used in this study was derived, has a long history of use of wettable sulphur in vineyards and it is possible that the predator has undergone selection for resistance during the past 60–80 years in which grapes have been grown in this region. The apparent universal low toxicity of synthetic fungicides to *T. doreenae* will also be a considerable advantage in developing biological control strategies using this predator.

Although fungicides appeared generally to be more harmful to *A. victoriensis*, only mancozeb, triadimefon, wettable sulphur and lime sulphur were highly toxic. Most other compounds permitted at least some survival, with benomyl, chlorothalonil, iprodione and procymidone relatively non-toxic. In a peach orchard trial, chlorothalonil had no impact on *A. victoriensis* while benomyl and iprodione significantly reduced but did not eradicate populations (James 1989).

Insecticides and miticides were generally highly toxic to *T. doreenae*, except for azinphos-methyl, carbaryl and malathion. An azinphos-methyl resistant strain of the related phytoseiid, *T. occidentalis*, is an important component of integrated mite management in pome fruit (Readshaw 1975, Hoy 1985), and *T. doreenae* may have the same predisposition for inheritance of this character. *A. victoriensis* was susceptible to all insecticides / miticides used in this study with only endosulfan permitting some survival at the field rate. James (1989) demonstrated survival of *A. victoriensis* following treatment with the miticides clofentezine, hexythiazox and propargite but none of these are used in viticulture.

Pesticides which cause high mortality to both predator species (lime sulphur, carbamult, chlorpyrifos, diazinon, dimethoate, methidathion) are unlikely to have a role in vineyard mite management programs based on biological control. *T. doreenae* is considered to be the most important predator in biological control of grapevine mites (James *et al.* 1995) and it is fortunate that this species appears to possess the greatest tolerance to viticultural pesticides. However, final assessment of the impact of pesticides on the survival and performance of *T. doreenae* must be done in the field. Sublethal effects of pesticides on predator biology should be examined. Some fungicides are known to affect reproduction and fecundity of phytoseiids (Ioriatti *et al.* 1992).

The results presented here accord well with current knowledge of incidence and abundance of *T. doreenae* and *A. victoriensis* in vineyards. In the Riverland of South Australia, disease control in grapevines is generally based on wettable sulphur and copper hydroxide and no synthetic insecticides are used. *T. doreenae* is the dominant phytoseiid (joined by *A. victoriensis* when sulphur sprays finish) and pest mites occur in small, non-damaging populations (James and Whitney 1993a). In the Canberra district, synthetic fungicides are often used yet *T. doreenae* still provides good mite control (James *et al.* 1995). The absence of *T. doreenae* in the Murrumbidgee Irrigation Area and Sunraysia (James and Whitney 1993a) may be linked to greater use of lime sulphur, carbamult and synthetic insecticides / miticides.

The apparent compatibility of a large number of pesticides, with survival of *T. doreenae* and *A. victoriensis* greatly enhances the prospects of widespread development and adoption of biological mite management programs in Australian viticulture.

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