SENSITIVITY OF *VENTURIA INAEQUALIS* AND *V. PIRINA* TO DODINE IN NEW ZEALAND

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ABSTRACT

The sensitivity of *Venturia inaequalis* and *V. pirina* to dodine was evaluated during the 1997/98 season using EC₅₀ and EC₉₅ values for colony growth on amended agar. Seventy one orchards with known fungicide application histories were sampled in the six main apple growing districts of New Zealand and *Venturia* isolates were tested. Sensitivity values were 19% lower in 1998 than indicated by data from 1994. No relationship between sensitivity and orchard fungicide spray history was detected. Despite its frequent use, dodine is still an effective fungicide for controlling apple black spot (*V. inaequalis*). Baseline sensitivities of *V. pirina* (pear black spot) for dodine were established from eight orchards in Hawkes Bay, Auckland and Central Otago. No dodine resistance was detected in this pathogen.

Keywords: *Venturia inaequalis*, *V. pirina*, fungicide resistance, reduced sensitivity, apple scab, pear scab.

INTRODUCTION

The pipfruit industry is one of New Zealand's main export earners, realizing \$ 354 million in 1997 (Luxton 1997). Apple and pear black spot (caused by *Venturia inaequalis* (Cooke) Wint. and *V. pirina* Aderh. respectively) are considered serious threats by most orchardists.

Thirty-three fungicide formulations are currently registered in New Zealand for the control of black spot in pip fruit. Dodine is a locally systemic protective fungicide with specific activity against black spot of apples, pears and roses with a 48 hr reach back (O'Connor 1998). Dodine has been used mainly against black spot on pome fruits, but owing to the costs and risks involved in developing new fungicides and an increase in the phenomenon of fungicide resistance, the possibility of using dodine against other diseases has been raised. In addition to having new applications, dodine may also be a useful substitute for products no longer used, and in anti-resistance strategies of mixtures or alternation with at-risk fungicides (Miglio 1994). The mechanism by which fungi acquire resistance to dodine is still unknown (Brent 1995).

The Plant Protection Research Unit established baseline sensitivities of *V. inaequalis* to dodine in a survey in 1994, to enable monitoring of possible shifts in pathogen populations with reduced sensitivity to the fungicide (Bakker *et al.* 1995). The current work is the first investigation after baseline sensitivities were established.

During preparations for sampling, some pear growers with concerns about the sensitivity of *V. pirina* in their orchards wished to be included and first sensitivity values of this pathogen to dodine are presented in this paper.

MATERIALS AND METHODS

Seventy one apple and pear orchards in Auckland, Waikato, Hawkes Bay, Nelson, Canterbury and Central Otago were sampled in December 1997 and January 1998. Use of dodine during the preceding two to three years was recorded to enable determination of correlation of use with fungicide sensitivities. Orchards were selected for sampling only when they had a known history of dodine use. Each orchard was sampled for a maximum of 1+ hours or until 15 fruit or leaves with black spot lesions were found.

Venturia spp. were isolated and tested as described previously (Bakker *et al.* 1995). Conidia were scraped from sporulating lesions, streaked on water agar (10 g/litre) and incubated for 2 to 3 days at $19\pm1^\circ\text{C}$ with a 12 h photoperiod of fluorescent light. Single germinated conidia were transferred to potato dextrose agar (PDA; (39 g/litre) amended with benzylpenicillin (100 ppm) and streptomycin sulphate (100 ppm) to reduce bacterial growth. Plates were incubated for 3 to 4 weeks at $19\pm1^\circ\text{C}$. Two hundred and eighty three isolates were obtained and maintained on PDA slopes. Six isolates per orchard were tested in a laboratory evaluation for sensitivity to dodine. Technical grade dodine (Nufarm NZ Ltd) was dissolved in ethanol and amended prior to autoclaving to a mixture of PDA (19.5 g/litre) and water agar (10 g/litre) at 0, 0.67, 1.33, 2, 2.67 and 5.33 mg/litre. Final ethanol concentrations in the medium were less than 1.5%. Benzylpenicillin and streptomycin sulphate (200 mg/litre each) were added to autoclaved medium. Mycelial plugs (4 mm diameter) from the actively growing colony margin of each isolate were transferred to three replicate plates containing agar amended with fungicide at each concentration. The plates were incubated at $19\pm1^\circ\text{C}$ darkness for 4 weeks, after which the diameters of resulting colonies were measured.

EC_{50} and EC_{95} values were calculated by linear regression of colony diameter with $\log_{10}$ fungicide concentration. EC_{95} values followed the pattern of the EC_{50} values and are therefore not presented in this paper. Resistance factors (Rf) were calculated relative to those of sensitive populations. EC_{50} values of isolates were correlated with the numbers of times that these isolates had been exposed to dodine as indicated by orchard fungicide spray histories.

RESULTS

Venturia inaequalis

The mean number of fruit or leaves with apple black spot lesions found in 1+ hours was 10.5 per orchard, with a maximum district mean of 13.7 in Canterbury and a minimum district mean of 6.3 in Nelson. Dodine use in each orchard varied from 0 to 15 applications per season, with the lowest mean number of applications per orchard in Auckland (0.8 per season) and the highest in Nelson (9.1 per season). The mean numbers of dodine applications of the orchards that were sampled were 3.3 in 1997/98 and 4.4 in 1996/97 and in 1995/96.

Mean dodine EC_{50} values for *V. inaequalis* varied from 0.114 mg/litre to 1.072 mg/litre, but district means varied only from 0.502 mg/litre in Hawkes Bay to 0.572 mg/litre in Central Otago (Table 1). These were lower than in 1994 (0.735 and 1.442 respectively) (Table 2).

TABLE 1: Mean, median, minimum and maximum dodine EC_{50} values (mg/litre) for *V. inaequalis* isolated in 1997/98.

District	Mean	Median	Minimum	Maximum
Auckland (5) ¹	0.505	0.473	0.336	0.816
Canterbury (11)	0.552	0.545	0.114	0.938
Hawkes Bay (14)	0.502	0.487	0.189	1.072
Nelson (16)	0.521	0.516	0.290	0.749
Central Otago (9)	0.572	0.554	0.368	0.806
Waikato (3)	0.537	0.551	0.348	0.846
National	0.527	-	0.114	1.072

¹Number of orchards from which isolates were tested

Resistance factors of *V. inaequalis* ranged from 0.551 to 2.028, but the mean district Rf values varied only from 1.129 to 1.183. For comparison with 1994 data, the Rf of the highest relative to the lowest EC_{50} was calculated (Table 2). The national mean Rf in 1994 was 1.96.

TABLE 2: Highest and lowest mean dodine EC₅₀ values (mg/litre) and calculated resistance factors (Rf) from the 1998 and 1994 tests.

	1998	1994
Highest	1.072	1.442
Lowest	0.114	0.735
Rf-value ¹	9.40	1.96

¹Rf-values are the highest values divided by the lowest values.

EC₅₀ values of the isolates were compared with the numbers of dodine applications made to the respective apple blocks, but no relationship was found although the high district mean was found in the district with the highest use.

Venturia pirina

Sensitivity of *V. pirina* ranged from 0.442 to 1.092 mg/litre (Table 3), while Rf values ranged from 0.84 to 1.15. This pathogen had not been monitored for dodine sensitivity before, but mean sensitivities were slightly higher than for *V. inaequalis*.

TABLE 3: Mean, median, minimum and maximum dodine EC₅₀ values (mg/litre) for *V. pirina* isolated in 1997/98.

District	Mean	Median	Minimum	Maximum
Auckland (2)	0.766	0.700	0.502	1.083
Hawkes Bay (3)	0.584	0.541	0.443	0.794
Central Otago (2)	0.743	0.766	0.442	1.092
National	0.690	-	0.442	1.092

To enable comparison of the Rf for *V. pirina* (2.47) with the Rf for *V. inaequalis* (9.40), the Rf of the highest value for *V. pirina* (1.092 mg/litre) was calculated relative to the lowest value of *V. pirina* (0.442 mg/litre).

DISCUSSION

Black spot samples were collected from blocks where a positive selection pressure for reduced sensitivity to dodine was expected by selecting orchards with dodine application histories during the last 2 to 3 years and by avoiding trees with obvious control failures other than that caused by fungicide resistance. However, no isolates were found with EC₅₀ values that suggested reduced sensitivity, and the national mean EC₅₀ for 1998 was 19% lower than that for 1994. A small reduction in numbers of spray applications due to weather conditions and fungicide resistance management strategies may have attributed to this.

The use of dodine in New Zealand apple orchards has reduced slightly over recent seasons. A mean number of applications of 3.3 in 1997/98, compared to 4.4 in the two previous years and 4.0 and 4.8 in 1993/94 and 92/93 respectively (Bakker *et al.* 1985, unpubl. data) probably reduced the risk of development of resistant pathogen populations. Dodine remains a viable option for apple and pear black spot control and could play an important role in management of resistance to Demethylation Inhibiting (DMI) fungicides. The present study has established baseline sensitivities of *V. pirina* to dodine and enables the monitoring for future shifts in fungicide sensitivity in this pathogen.

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