MEASUREMENT OF SENSITIVITY TO DMI FUNGICIDES IN POPULATIONS OF *UNCINULA NECATOR*

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SUMMARY

Isolates of *Uncinula necator* (Schw.) Burrill, which causes powdery mildew of grapes, were collected from the main wine grape growing areas. A method is described for the multiplication of the pathogen on detached leaves and leaf discs of susceptible grape cultivars. Inoculum from such multiplications was tested on leaf discs treated with triadimenol as a marker for sensitivity to other related DMI fungicides. A discriminating dose of 0.30 mg/litre was found to be satisfactory for preliminary screening on 'Chardonnay' leaves. Twenty isolates tested were considered to be sensitive to triadimenol. Further testing with more isolates and different DMI products is required to determine whether this is representative for *U. necator* in New Zealand.

Keywords: *Uncinula necator*, grapes, triadimenol, sensitivity evaluation

INTRODUCTION

Powdery mildew, caused by *Uncinula necator* (Schw.) Burrill, is a serious disease in all grape producing areas in the world. The fungus infects both the leaves and the berries. The disease is considered to be more important in table grapes than in wine grapes, because it reduces fruit quality and marketable yield as well as total yield. Heavy infections in wine grapes may result in large yield losses because of reduced berry harvest and the potential to affect wine quality.

The demethylation inhibitor (DMI) fungicides have been used in many grape growing countries for efficient control of powdery mildew. Cyproconazole, fenarimol, myclobutanil, triforine, triadimefon and penconazole have label claims for use on grapes in New Zealand and there are recommendations from the New Zealand Committee for Pesticide Resistance (NZCPR) for restrictions in the total number of DMI applications in each season (Prince *et al* 1989). These restrictions were recommended because of concerns about the possible development of resistance to the DMI fungicides. In 1988, reduced efficiency in control of grape powdery mildew was reported from Portugal (Steva *et al* 1988) and it was suspected that strains of *U. necator* with reduced sensitivity had developed. Since then, strains with reduced sensitivity to several DMI fungicides have been reported in France (Steva and Clerjeau 1990), Italy (Aloi *et al* 1990), California and most recently in Israel (Thind *et al* 1992).

In this paper we describe a modified method for sensitivity evaluation of powdery mildew populations, and report the results of preliminary tests on twenty isolates collected from the major wine grape production areas in New Zealand.

MATERIALS AND METHODS

Grape leaves and berries infected with powdery mildew were collected during the summer (December 1991-March 1992). More than 30 vineyards were visited and *U. necator* population samples were obtained from 20 vineyards for multiplication in the laboratory and for fungicide sensitivity tests.

Leaf and disc preparations

Whole, detached young leaves of *Vitis vinifera* L. cvs 'Chardonnay', 'Blauberger' and 'Gaillard Gerard' were used to produce conidia and maintain the pathogen (Steva *et al* 1992). The leaves were obtained from plants untreated with DMI fungicides from the Lincoln University vineyard from December 1991- February 1992. Cuttings,

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planted in 200 mm plastic bags in December 1991 and grown in a glasshouse, supplied young leaves during March-April 1992. The plants and cuttings were treated prophylactically with sulphur (Super Six, Elliott Chemicals Ltd) to prevent infection. The leaves were washed twice in tap water to remove sulphur residues, and surface sterilized for 5 minutes in a solution of calcium hypochloride (100 g/litre of 35% active chlorine). The leaves were rinsed twice in sterile water and placed between sterile paper. They were then transferred adaxial surface uppermost for inoculation and incubation, or were used to cut leaf discs with a 18 mm sterile cork borer for fungicide assays.

Fungicide assays

Triadimenol (Cereous 250 g/litre EC, Bayer NZ Ltd) was used for sensitivity assays. Leaf discs were placed in 85 mm petri dishes, with the adaxial surface down, on 80 mm filter paper saturated with fungicide suspension or with sterile water. Each dish contained 10 discs cut from different leaves, as replicates for each treatment. In most tests four discs were obtained from each leaf, and used for one water check and three fungicide concentration (0.3, 1 and 3 mg/litre) treatments. Leaf discs were left for 24 h in contact with the water or fungicide, and then transferred with the adaxial surface uppermost for inoculation and incubation.

Whole leaves and leaf discs were placed in 85 mm petri dishes containing water agar amended with benzimidazole (30 mg/litre). A 82 mm disc of filter paper was placed to cover the medium, saturated with 2 ml sterile water, and the leaves or leaf discs were placed on top of the wet filter paper. The wet adaxial surface was allowed to dry before the discs were inoculated. Conidia from each sample were dislodged with a fine airstream and allowed to settle in a perspex tower (400 mm high) onto the adaxial surface of whole leaves. In each fungicide test four petri dishes, each containing 10 discs for one treatment, were placed in a settling tower (200 x 200 x 400 mm high) and inoculated simultaneously. The dishes were then closed and placed in a growth room at 20°C with 12 h of fluorescent light per day. All operations were carried out in a laminar flow cabinet to retain sterility in the experiments. Disease and pathogen development were observed microscopically from 5 days after the inoculations for a further 12 days or more. The latent period (first conidia produced) and incubation periods (visually apparent powdery mildew spots) were determined.

RESULTS AND DISCUSSION

The cultivar 'Chardonnay' was found to be most suitable for pathogen multiplication and fungicide testing. Pathogen establishment occurred readily in the test conditions, and sporulation was detectable after 7 to 10 days, depending on the inoculum concentration applied. Development was slower and more erratic on 'Blauberger' and 'Gaillard Gerard', but these were used occasionally for multiplication of inoculum. Preliminary tests established that several isolates from Canterbury vineyards and from a Christchurch home garden were totally inhibited from colonising and sporulating on detached leaf discs treated with 0.3 mg/litre triadimenol. There was limited growth on discs treated with 0.15 mg/litre and sparse sporulation on discs treated with 0.075 mg/litre. Subsequent tests were conducted on discs treated with 0, 0.3, and 1.0 mg/litre triadimenol. Any isolates with any indication of growth or sporulation on discs treated with 0.3 mg/litre were retested on the above range plus 0.15 mg/litre triadimenol.

Five Canterbury isolates, four Marlborough isolates, three Nelson isolates, two isolates from Otago, Martinborough and Gisborne, and one isolate from Auckland and Hawkes Bay vineyards were totally inhibited by 0.3 mg/litre triadimenol. One isolate from each of Canterbury, Marlborough and Martinborough were retested because there was some development in the initial test, but there was no sporulation on discs treated with 0.3 or 1.0 mg/litre in the confirmation tests. A preliminary test with other DMI products indicated that a sub-sample of isolates were inhibited at similar concentrations of fungicides.

Isolates were derived where possible from vineyards with a history of regular use of DMI products. There was no detectable shift in sensitivity to DMI fungicides in the powdery mildew populations, although the sample size was small and not all

products were tested on all isolates. Triadimenol is a useful marker for resistance to this group, but cross-resistance is not always present. Further monitoring should be conducted on a regular basis, including the establishment of baseline EC₅₀ values for the most widely used products.

Growers reported some concern over the widespread occurrence of powdery mildew, but there is little documentation of the losses incurred from reduced harvest or wine quality. The current growth of the wine industry, particularly in the drier areas such as Canterbury, Marlborough and Otago and with the cvs 'Chardonnay', 'Sauvignon Blanc' and 'Pinot Noir' which are all susceptible to powdery mildew, suggests that the disease may become more prevalent and damaging. If so, pressure for the development of resistance may increase by greater chemical usage, which should be treated with great caution. Most growers appeared to be adhering to the recommendations of the NZCPR (Prince *et al* 1989), although five applications had been made on a few properties in some years, and there was less combination with sulphur than recommended. Continued implementation of the recommendations and periodic checks may delay or prevent the development of resistance as has occurred in Europe.

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