Impairment of grapevine xylem function by *Phaeomoniella* chlamydospora infection is due to more than physical blockage of vessels with 'goo'

JACQUELINE EDWARDS, IAN G. PASCOE and SOHEIR SALIB

Primary Industries Research Victoria, Knoxfield Centre, Private Bag 15, Ferntree Gully Delivery Centre, Victoria 3156, Australia

Summary. *Phaeomoniella chlamydospora* is a vascular pathogen that colonises the woody xylem tissues of the grapevine. It is associated with the grapevine trunk diseases, esca and Petri disease. Infection is usually accompanied by a dark tarry substance, commonly refered to as 'black goo', in some of the xylem vessels. Examination of field grown Verdelho demonstrated that infection reduced xylem function by 16% for each 1% increase in 'goo'-blocked vessels, indicating that vessel blockage is not solely responsible for loss of xylem function.

Key words: vascular pathogen, esca, Petri disease, grapevine trunk disease.

Introduction

Grapevine trunk diseases limit the long-term sustainability of winegrape production in Australia. They affect the viability of newly planted vineyards and inhibit the productivity of, and in many cases kill, mature vines as they reach an elite stage of maturity. The grapevine trunk diseases, Petri disease, and esca are caused by the xylem-inhabiting fungal pathogen, *Phaeomoniella chlamydospora*, although other fungi have been implicated (Crous and Gams, 2000). Of the two diseases, Petri disease is the more prevalent in Australia (Edwards and Pascoe, 2004). The disease primarily affects young grapevines, and symptoms include graft failure, shoot dieback, slow decline and gradual death

Corresponding author: J. Edwards

E-mail: jacky.edwards@dpi.vic.gov.au

of the grapevine (Ferreira *et al.*, 1994). The disease has a characteristic symptom of internal black wood streaking, evident when the trunk of an infected grapevine is cut open to reveal black tarry 'goo' in the affected xylem vessels. Ferreira *et al.* (1994) suggested that the disease symptoms were directly attributable to this blockage of xylem vessels. The research presented here aimed to investigate whether impairment of xylem function could be correlated directly with 'goo-blocked' vessels.

Materials and methods

The trial site was a vineyard, cv. Verdelho, in the Yarra Valley, Victoria, planted in 1998, which had been diagnosed with Petri disease when the vines were 3 years old. In April 2002, a few days prior to harvest, 10 vines were cut at the base of the trunk, placed in buckets of dilute methylene blue solution (5 g in 25 l water), and left in the field for 24 h, based on a method described by Tarara and Ferguson (2001). On collection, the trunks

Fax: +61 3 9800 3521.

were sectioned into 10-cm pieces, and % functional (i.e. blue), non-functional (i.e. white) and 'gooblocked' vessels were calculated for each piece using image analysis software and then combined to give estimates for each grapevine. The experiment was repeated in April 2004 with five grapevines from the same block. A linear regression trendline was fitted using MS Excel to describe the relationship between the percentage of functional and gooblocked' vessels.

Results

Within a few hours, the blue dye had been translocated right throughout the grapevines and was easily visible at the shoot tips and tendrils. The grapevines were in full canopy and ready for harvest, and the weather was warm (> 35° C in 2002 and 27^{\circ}C in 2004). When the grapevine trunks were recovered after 24 hours, it was immediately apparent that the more 'black goo' in the trunk, the less blue dye had been translocated (Fig. 1). A strong negative relationship was evident between the percentage of functional vessels and those blocked with black goo. A similar relationship was seen for data from both 2002 and 2004, so that the two data sets were combined (Fig. 2). For the combined data set, the relationship could be described by the equation y = -15.904 x + 77.659(R²=0.7247) where y=% functional xylem vessels and x=% vessels blocked with black goo.

Discussion

The results presented here clearly showed that *P. chlamydospora* infection in field-grown Verdelho considerably reduced xylem function. Vascular pathogens are reported to block xylem vessels by induction of tyloses, production of gums (such as black goo) or with physical structures of the pathogen, and Ferreira *et al.* (1994) concluded that this was responsible for the symptoms expressed in Petri disease (then known as slow dieback of grapevines). Grapevines have a very efficient vascular

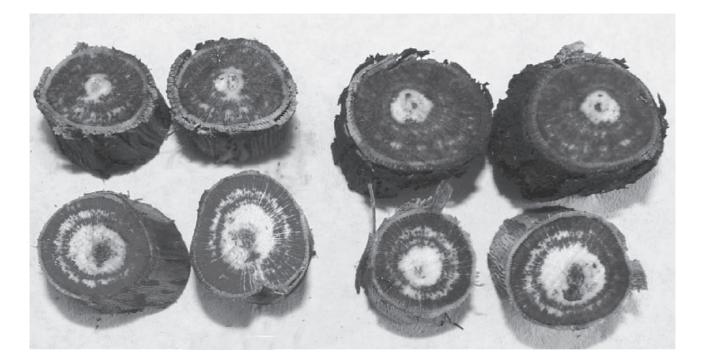


Fig. 1. Comparison of the functional xylem (blue coloured vessels) of two five-year-old Verdleho grapevines. Top, trunk of a grapevine with no black goo, 76% functional xylem. Bottom, trunk of a grapevine with 2% vessels blocked with black goo, 54% functional xylem.

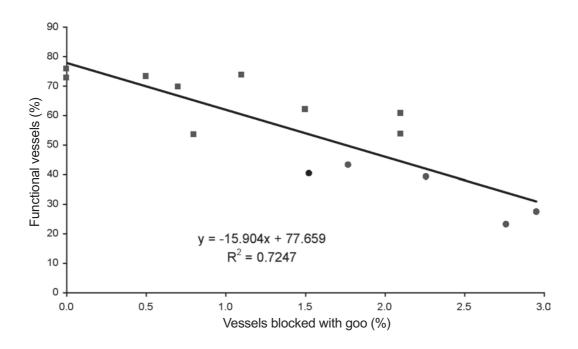


Fig. 2. Relationship between percentage of functional xylem and percentage of xylem blocked with black goo of Verdelho grapevines infected with *P. chlamydospora*. \blacksquare = 2002 data, \blacklozenge = 2004 data.

system, however, requiring only 2–3 annual increments of functional vascular tissue to sustain vigorous shoot growth and heavy fruiting (Pratt, 1974; Mullins *et al.*, 1992). In the present study, it was apparent that *P. chlamydospora* infection interfered with xylem function (as expected for a vascular pathogen), but that the interference was due to more than physical blockage of vessels. The relationship using field-grown Verdelho showed that for each 1% increment of xylem blocked with goo, there was a corresponding loss in functional xylem of approximately 16%.

Over the past few decades, research has demonstrated that vascular pathogens can interfere with xylem function in more ways than occlusion (Ayres, 1978; Zimmerman, 1983; Pegg, 1985). Van Alfen *et al.* (1983) hypothesised that macromolecules produced by vascular pathogens plugged pit membranes, thus reducing translocation of water. Tyree and Sperry (1989) speculated that production of oxalic acid by pathogens alters the permeability of pit membranes, causing cavitation to occur. Studies of the vascular disease Dutch elm disease clearly demonstrated that the pathogen, *Ceratocystis ulmi*, was responsible for air-seeding and inducing embolisms in vessels quite removed from the site of infection (Newbanks *et al.*, 1983). More detailed research than that of the present study is required, however, to determine the mechanism(s) by which *P. chlamydospora* disrupts grapevine xylem function.

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References

Ayres P.G., 1978. Water relations of diseased plants. In: Water Deficits and Plant Growth. Vol 5. (T.T. Kozlowski, ed.), Academic Press, London, United Kingdom, 1–60.

- Bertelli E., L. Mugnai and G. Surico, 1998. Presence of *Phaeoacremonium chlamydosporum* in apparently healthy rooted grapevine cuttings. *Phytopathologia Mediterranea* 37, 790–82.
- Crous P.W. and W. Gams, 2000. *Phaeomoniella chlamydospora* gen. et comb. nov., a causal organism of Petri grapevine decline and esca. *Phytopathologia Mediterranea* 39, 112–118.
- Edwards J., G. Marchi and I.G. Pascoe, 2001. Young esca in Australia. *Phytopathologia Mediterranea* 40, 303– 310.
- Edwards J. and I.G. Pascoe, 2004. Occurrence of *Phaeomo*niella chlamydospora and *Phaeoacremonium ale*ophilum associated with Petri disease and esca in Australian grapevines. Australasian Plant Pathology 33, 273–279.
- Ferreira J.H.S., P.S. van Wyk and F.J. Calitz, 1999. Slow dieback of grapevine in South Africa: stress-related predisposition of young vines for infection by *Phaeoacre*monium chlamydosporum. South African Journal of Enology and Viticulture 20, 43–46.
- Ferreira J.H.S., P.S. van Wyk and E. Venter, 1994. Slow dieback of grapevine: association of *Phialophora parasitica* with slow dieback of grapevines. *South African Journal of Enology and Viticulture* 15, 9–11.
- Fourie P.H. and F. Halleen, 2004. Proactive control of Petri disease of grapevine through treatment of propagation material. *Plant Disease* 88, 1241–1245
- Gubler W.D., K. Baumgartner, G.T. Browne, A. Eskalen, S. Rooney-Latham, E. Petit and L.A. Bayramian, 2004. Root diseases of grapevines in California and their control. Australasian Plant Pathology 33, 157–165.

- Gubler W.D., P.E. Rolshausen, F.P. Trouillas, G.M. Leavitt and E.A. Weber, 2006. Grapevine trunk diseases in California. In: Paper and Abstracts Book, 6th International Cool Climate Symposium for Viticulture and Oenology, 6–10 February 2006, Christchurch, NZ, 121–125.
- Halleen F., P.W. Crous and O. Petrini, 2003. Fungi associated with healthy grapevine cuttings in nurseries, with special reference to pathogens involved in the decline of young vines. *Australasian Plant Pathology* 32, 47– 52.
- Mullins M.G., A. Bouquet and L.E. Williams, 1992. *Biology* of the Grapevine, Cambridge University Press, United Kingdom, 52–54.
- Newbanks D., A. Bosch and M.H. Zimmerman, 1983. Evidence for xylem dysfunction by embolization in Dutch elm disease. *Phytopathology* 73, 1060–1063.
- Pegg G.F., 1985. Life in a black hole the micro-environment of the vascular pathogen. *Transactions of the British Mycological Society* 85, 1–20.
- Pratt C., 1974. Vegetative anatomy of cultivated grapes a review. American Journal of Enology and Viticulture 25, 131–149.
- Tarara J.M. and J.C. Ferguson, 2001. Device for simulating high rates of sap flow in grapevines. *American Journal of Enology and Viticulture* 52, 260–265.
- Tyree M.T. and J.S. Sperry, 1989. Vulnerability of xylem to cavitation and embolism. *Annual Review of Plant Physiology and Molecular Biology* 40, 19–38.
- Van Alfen N.K., B.D. McMillan, V. Turner and W.M. Hess, 1983. Role of pit membranes in macromolecule-induced wilt of plants. *Plant Physiology* 73, 1020–1023.
- Zimmerman M.H., 1983. Xylem Structure and the Ascent of Sap. Springer-Verlag, Berlin, Germany, 139 pp.

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