



# First report of collar and root rot caused by *Phytophthora hedraiaandra* on *Viburnum* in the UK

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*Viburnum* is a popular shrub grown mainly for its flowers, fragrance, berries, autumn colour, and it is also used as a hedge. In 2011, samples of *Viburnum* showing symptoms of collar and root rot collected in North Yorkshire (RHS166247) were received by the advisory service at the Royal Horticultural Society. The plant was part of a hedge showing signs of severe dieback with many plants dying. A *Phytophthora* species was isolated from root samples using rhododendron leaves for baiting. Freshly picked leaves of *Rhododendron catawbiense* 'Cunningham's White' were washed under running tap water and blotted dry on filter paper. The roots were placed in a plastic tray and flooded with filtered pond water and the intact rhododendron leaves were floated on top of the water. The trays were left at room temperature (18–25°C) on the bench. After two to eight days, 5 mm<sup>2</sup> sections were aseptically removed from the margins of developing lesions, plated onto P5ARP (Erwin & Ribeiro, 1996) and incubated at 20°C in the dark. Single hyphal tip cultures were obtained by transferring individual hyphal tips from the P5ARP plates onto carrot agar (CA) (Erwin & Ribeiro, 1996). Sporangia were produced and zoospores released in pond water following floating of CA plugs.

*Phytophthora hedraiaandra* RHS166247 was identified using morphological characteristics and confirmed by DNA sequencing. Sporangia were conspicuously papillate (av. 2.3 µm) and were ovoid to obpyriform, caducous with average dimensions of 37.7 ± 1.1 x 26.1 ± 0.8 µm (average l/b ratio 1.45) and short pedicels (average 1.2 µm). The gametangia were readily produced on CA within 7 days. The fungus was homothallic and the antheridia were paragynous occasionally amphigynous. Oogonia had smooth walls and were usually globose to slightly subglobose. They had a mean diameter of 36.0 ± 0.5 µm. Oospores were slightly aplerotic and measured 29.2 ± 0.5 µm. The oospore wall index was 0.22. Antheridia measured 10.5 ± 0.3 x 9.9 ± 0.6 µm. The morphology of sporangia and gametangia agreed with the original description of *Phytophthora hedraiaandra* by De Cock & Lévesque (2004). The identification of the isolate RHS166247 was confirmed by direct sequencing of the ITS region using a semi-nested PCR reaction. The primers ITS4 (White *et al.*, 1990) and DC6 (Bonants *et al.*, 1997) were used in the first round and ITS4 and ITS6 (Cooke *et al.*, 2000) in the second round. The PCR conditions for both rounds were the same as those described by Cooke *et al.* (2000) except for the annealing temperature which was 62°C in the first round and 63°C in the second round. Sequencing of the ITS region revealed that the nucleotide sequence of the isolate RHS166247 (GenBank Accession No. KF793827) is 99% identical to the ex-type culture of *Phytophthora hedraiaandra* CBS111725 (AY707987). The ITS sequence also has three polymorphisms at positions 73, 99 and 100 of the ITS1 which differentiates

it from its close relative *P. cactorum*.

Six plants of *Viburnum tinus* were stem inoculated with 3 mm plugs from a seven-day-old *Phytophthora* culture (RHS166247) grown on CA. A bark incision was made using a sterile 3 mm cork borer to expose the cambium and the plug placed in the incision. The wounds were wrapped in damp sterile cotton wool, Parafilm and foil. The plants were kept in a polycarbonate grow dome where the temperature was maintained at 20°C during the day and at 15°C during the night. *P. hedraiaandra* was pathogenic to *Viburnum tinus* causing necrotic phloem lesions 15 days after inoculation, with average lengths of 35.9 ± 6.0 mm whereas the agar control produced lesions of 11.5 ± 2.3 mm. *P. hedraiaandra* was successfully re-isolated from the margins of the lesions of the inoculated plants but not from the controls. *P. hedraiaandra* occurs on *Rhododendron* and *Viburnum* species in Italy, Spain, Slovenia, the Netherlands and the USA (Farr *et al.*, 2013). It causes wilting, stem cankers and root and collar rots. To our knowledge, this is the first finding of this species in the UK. Following its detection another case in a garden in London was confirmed by direct sequencing of plant material taken at the collar of *Viburnum tinus* (GenBank Accession No. KF793828). No statutory action is taken against this species.

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Figure 1



Figure 2

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