



## *Thuja occidentalis*: a new host for *Phytophthora lateralis*

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In February and March 2011, Scottish Government Plant Health Inspectors submitted three potted *Thuja occidentalis* cv. 'Emeraude' plants to SASA requesting a test for the presence of *Phytophthora*. They were collected in a nursery in central Scotland, from a batch of 60 plants about one metre in size originally imported from France, and were showing a grey-olive discolouration on parts of the foliage with dieback beginning on some branches (Figs. 1, 2). No symptoms were observed on the bark, root collar or roots. Foliage and roots were tested separately in a nested PCR described by Schena *et al.* (2008) targeting the Ras-related protein gene *Ypt1*. The first round of this PCR with general *Phytophthora* primers YPh 1F/2R yielded an amplification product of ~ 470 bp. The PCR product was then amplified again in a second PCR with specific *P. lateralis* primers Ylat3F/2R resulting in a band matching the positive control extracted from a pure culture of *P. lateralis* (133 bp). The positive result was obtained only from the foliage samples; the roots were tested with negative result indicating an aerial rather than root infection, as already observed by Robin *et al.* (2011).

Parallel with the PCR test, small (approx. 5 mm) discoloured leaflets had been plated onto V8 agar supplemented with antibiotics (PARPNH; Jung *et al.*, 1996) and incubated at 18°C in the dark. A slow-growing *Phytophthora* sp. was observed after five days that was transferred onto V8 agar without antibiotics. The culture grew submerged with sparse aerial mycelium, producing non-caducous, non-papillate sporangia, generally ovoid but often distorted. The typical laterally formed chlamydospores of *P. lateralis* could be observed after 7-10 days (Fig. 3). No oospores were observed from the isolate grown on V8 agar. The ITS region of the ribosomal RNA gene and the *Ypt1* gene of the isolate were sequenced (GenBank Accession Nos. JN182996, JN182997) and confirmed the identity of *P. lateralis*. The closest matches (99% identity) were GenBank accessions FJ196746 for the ITS sequence and DQ162991 for *Ypt1*.

Pathogenicity of the isolated culture was tested on *Thuja occidentalis* cvs. 'Smaragd' and 'Holmstrup' and *Chamaecyparis lawsoniana* cvs. 'Ellwoodii' and 'White Spot'. Ten leaflets (7-10 mm) per host were floated in a petri dish containing sporangia of the isolated *P. lateralis* (~3500 sporangia/ml).

For the negative control another 10 leaflets from the same hosts were floated in sterile distilled water. The petri dishes were kept in a growth room at 20°C with 12 hours light. After seven days, most leaflets on all tested hosts floating in the sporangia suspension were showing discolouration or necrosis. The leaflets were surface-sterilised, plated onto V8 agar with antibiotics as before and the pathogen was re-isolated, completing Koch's postulates. The only confirmed hosts for *P. lateralis* have been *Chamaecyparis lawsoniana* and *Taxus brevifolia* (Hansen & Lewis, 1997). This is the first report on *Thuja occidentalis*. Infected and surrounding plants in the nursery were destroyed to eradicate the disease.

### References

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Figure 1



Figure 2



Figure 3

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