Project title: Evaluation of fungicides and novel treatments for the control of black root rot, *Thielaviopsis basicola*, in bedding and hardy nursery stock plants

Project number: HNS-PO 190

Project leader: Dr Erika F. Wedgwood, ADAS

Report: Final report, August 2017

Previous report: August 2014, 2015 and 2016

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2015 to 2017 Choisya: Fletchers Lane site of New Place Nurseries Ltd, Sidlesham Common, West Sussex, PO20 7QG

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Date project commenced: 1st September 2013

Date project completed (or expected completion date): 31st October 2017
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The results and conclusions in this report are based on an investigation conducted within a one-year period. The conditions under which the experiments were carried out and the results have been reported in detail and with accuracy. However, because of the biological nature of the work it must be borne in mind that different circumstances and conditions could produce different results. Therefore, care must be taken with interpretation of the results, especially if they are used as the basis for commercial product recommendations.
AUTHENTICATION

We declare that this work was done under our supervision according to the procedures described herein and that the report represents a true and accurate record of the results obtained.

Dr Erika F. Wedgwood
Research Scientist
ADAS UK Ltd

Signature E. F. Wedgwood Date 27 October 2017

Report authorised by:
Dr Barry Mulholland
Head of Horticulture
ADAS UK Ltd

Signature Barry J. Mulholland Date 27 October 2017
GROWER SUMMARY

Headline
- Black root rot severity in Viola sp. was reduced by Cercobin WG, Signum, three coded fungicides and one coded biofungicide; it appeared to be lessened by Prestop and another coded biopesticide.
- Neither a single application of Cercobin WG nor three applications of biofungicides (various products and sequences) reduced black root rot or increased plant vigour of Choisya.
- Infection by Thielaviopsis basicola may be underestimated; symptoms are fully described and illustrated in this report.
- Fusarium oxysporum was isolated from roots of wilted and dying Choisya; further work is required to determine if this a cause of Choisya wilt.

Background
Symptoms and prevalence
Black root rot (Thielaviopsis basicola, syn. Chalara elegans) affects plants from at least 15 families, causes root damage leading to reduced nutrient and water uptake, consequent leaf yellowing and potentially plant death. Pigmented resting spores (chlamydospores) form in roots and when abundant cause blackening of sections of root (Figure 1). Losses to black root rot in Viola spp. production can be substantial, and some hardy nursery stock species such as Choisya, Skimmia and Ilex are particularly susceptible. Black root rot is a long-established problem for growers, and in the UK, it is likely that around 15% of ornamentals production on nurseries is susceptible to black root rot. It has been estimated that in any year 25% of susceptible crops on UK nurseries can be affected by the disease, with a fifth of these failing to be sold because either the reduction in quality is too great or the infection has been so severe that the plants have died.

Figure 1:
Low magnification of natural infection of Viola sp. roots by T. basicola.

Brown chlamydospore clusters inside the outer cells of roots causing lengths of root to appear blackened, or in lower numbers producing dark specks on whiter roots.
In bedding plants, such as *Viola* and *Primula* species, losses occur on nurseries within the couple of months in which the crops are being grown up to flowering. It is probable that around 2% of bedding and pot plants of these species become affected annually by black root rot, but as growers are aware of these plants’ susceptibility, fungicide treatments can reduce this loss to around 0.5% of total plants being unmarketable. Losses tend to be greatest in July as the plants can suffer from heat stress and become more susceptible to infection. In nursery stock species, such as *Choisya* and *Skimmia*, losses are often seen during establishment shortly after crops are potted up, whereas losses tend to be seen in finished plants of herbaceous species such as geraniums. In addition to losses facilitated by stress from heat or root disturbance, early crops can succumb to black root rot when conditions are cold.

**Cultural control**
Growers aim to employ cultural control measures such as reducing plant stress and taking care over crop hygiene to reduce the chance of plants becoming infected. However, the pathogen produces resting spores (chlamydospores) in roots which then survive in debris in matting, re-used containers and soil, and can be resistant to disinfectant treatments (as reported in AHDB Project PC 38c and Factsheet 03/14, revision 15/05). Details of various measures were given in the review of black root, PO 14 (Wedgwood, 2013).

**Biological control**
The biological products Prestop (*Gliocladium catenulatum*), Serenade ASO (*Bacillus subtilis*), T34 Biocontrol (*Trichoderma asperellum*), Trianum-G and Trianum-P (both *Trichoderma harzianum* T-22) can be used on ornamentals in the UK against root rots principally targeting *Pythium* and *Phytophthora* spp. although Prestop product information also lists *T. basicola*. The effect of either Serenade ASO, T34 Biocontrol, Trianum-G or Trianum-P against black root rot is unclear.

**Chemical control**
Growers of crops susceptible to black root rot usually treat them with a protectant fungicide drench. Cercobin WG (thiophanate-methyl) is often applied to container plants at sowing (bedding plants) or potting-on (nursery stock). The resistance risk with this benzimidazole fungicide is high. This product also protects against *Cylindrocarpon*, *Rhizoctonia* and *Fusarium* species. Treatment with products such as Subdue (metalaxyl-M) or Fenomenal (fenamidone + fosetyl-aluminium) are used in addition against the oomycete pathogen species of *Pythium* and *Phytophthora* but do not control *T. basicola* sp.. Cercobin WG can only be applied to container grown ornamentals under permanent protection, once per crop, and plants must be left for three weeks before planting into open ground (EAMU 2011 1887).
One application can be inadequate to maintain protection for nursery stock under long production seasons.

Summary

This project has sought to identify novel drench treatments with potential preventative or curative efficacy against black root rot, in order to seek use to supplement the small number of products available for use on pot and bedding plants and hardy nursery stock. The treatments (Table 1) included conventional chemicals and chemical elicitors and microbial products. The elicitors were withdrawn from testing after the second year due to registration issues.

Table 1. Products tested in the current project on either Viola sp. and/or Choisya sp..

<table>
<thead>
<tr>
<th>Product or experimental code</th>
<th>Active ingredient</th>
<th>Mode of Action</th>
<th>Fungicide group or product type</th>
<th>FRAC code</th>
<th>Permitted use in UK on ornamentals</th>
<th>Approval status and additional comments</th>
</tr>
</thead>
<tbody>
<tr>
<td>Conventional chemical fungicides</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Cercobin WG</td>
<td>thiophanate-methyl</td>
<td>Systemic</td>
<td>MBC or benzimidazole</td>
<td>1</td>
<td>Yes</td>
<td>Yes</td>
</tr>
<tr>
<td>HDC F173 Confidential</td>
<td></td>
<td></td>
<td>SDHI</td>
<td></td>
<td>No</td>
<td>No</td>
</tr>
<tr>
<td>Signum</td>
<td>bosalid + pyraclo-strobilin</td>
<td>Systemic &amp; protectant</td>
<td>SDHI + Qol</td>
<td>7 + 11</td>
<td>Yes</td>
<td>Yes</td>
</tr>
<tr>
<td>Switch</td>
<td>cyprodinil + fludioxonil</td>
<td>Systemic &amp; protectant</td>
<td>Anilino-pyrimidine + phenylpyrole</td>
<td>9 + 12</td>
<td>Yes</td>
<td>Yes</td>
</tr>
<tr>
<td>HDC F174 Confidential</td>
<td>Protectant (some curative)</td>
<td>DMI Qol</td>
<td>-</td>
<td>No</td>
<td>No</td>
<td>Approved product only on other crops</td>
</tr>
<tr>
<td>HDC F175 Confidential</td>
<td></td>
<td></td>
<td>Experimantal</td>
<td></td>
<td>No</td>
<td>No</td>
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<tr>
<td>HDC F176 Confidential</td>
<td>Systemic &amp; protectant</td>
<td>DMI Qol</td>
<td>3 + 11</td>
<td>No</td>
<td>No</td>
<td>Approved product only on other crops</td>
</tr>
<tr>
<td>Biofungicides and other biological products</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Prestop</td>
<td>Gliocladium catenulatum J1446</td>
<td>Protectant microbial</td>
<td>fungus</td>
<td>-</td>
<td>No</td>
<td>Approved on protected ornamentals</td>
</tr>
<tr>
<td>Serenade ASO</td>
<td>Bacillus subtilis QST 713</td>
<td>Protectant microbial</td>
<td>bacteria</td>
<td>44</td>
<td>Yes</td>
<td>Yes EAMU 0708 of 2013 for ornamentals</td>
</tr>
<tr>
<td>T34 Biocontrol</td>
<td>Trichoderma asperellum T34</td>
<td>Protectant microbial</td>
<td>fungus</td>
<td>-</td>
<td>No</td>
<td>Yes EAMU full protection 1118 of 2012</td>
</tr>
<tr>
<td>Trianum -G HDC F177</td>
<td>Trichoderma harzianum T22</td>
<td>Protectant microbial</td>
<td>fungus</td>
<td>-</td>
<td>No</td>
<td>Full Approval for fully protected plants</td>
</tr>
<tr>
<td>Trianum-P HDC F190</td>
<td>Trichoderma harzianum T22</td>
<td>Protectant microbial</td>
<td>fungus</td>
<td>-</td>
<td>Yes</td>
<td>Full Approval for fully protected plants</td>
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<tr>
<td>Horti-Phyte</td>
<td>Potassium phosphate</td>
<td>Nutrient &amp; stimulant</td>
<td>chemical</td>
<td>-</td>
<td>Yes</td>
<td>Liquid fertiliser</td>
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<tr>
<td>HDC F178 Confidential</td>
<td>Plant activator</td>
<td></td>
<td>chemical</td>
<td>-</td>
<td>No</td>
<td>No Not approved in the UK</td>
</tr>
<tr>
<td>HDC F179 Confidential</td>
<td>Protectant microbial</td>
<td></td>
<td>fungi</td>
<td>-</td>
<td>No</td>
<td>No Not approved in the UK</td>
</tr>
</tbody>
</table>
Dose rates and water volumes were as on the product labels, or as advised by their product technical managers as being most suitable in order to test what might be permitted in an Extension of Authorisation for Minor Use (EAMU). The standard was a Cercobin WG drench, with untreated uninoculated and untreated inoculated plants as controls.

**Objective 1:** To determine the efficacy against black root rot and plant safety of some chemical plant protection products to *Viola* sp. (Experiment 1; 2014).

**Objective 2:** To determine the efficacy against black root rot and plant safety of some biological plant protection products and plant stimulants to *Viola* sp. (Experiment 2; 2014).

*Viola* sp. were sown in 2014 in multicell trays at ADAS Boxworth. Drench or overhead spray treatments were made before and after spore drenches of *T. basicola* four weeks after sowing. Roots were examined microscopically at termination in order to confirm infestation. Three out of the four experimental products (HDC F174, F175 and F176) very significantly reduced root rot severity on *Viola* sp. whether applied preventatively alone or followed by curative application. Preventative application of Signum (but not Switch) was also effective. Control by these products was equivalent to that given by Cercobin WG where 17.7% of the root surface was brown in contrast to 35.5% in the untreated inoculated plants. The plant activator HDC F178 applied both preventatively and curatively significantly reduced root rot severity in *Viola* sp. to 6.3% compared with 18.0% in the untreated inoculated. Both Trianum-G incorporation pre-sowing and Prestop applied preventatively plus curatively very significantly increased the proportion of *Viola* sp. plugs without root rot, with a mean 38% healthy in contrast to 0% following Cercobin WG application.

**Objective 3:** To determine the efficacy of simple programmes of products found to be effective in controlling black root rot in *Viola* spp. in Experiments 1 and 2. (Experiment 3; 2014)

Product selection for Experiment 3 was based on the results of foliar vigour at intervals throughout the nine or 10 weeks of Experiments 1 and 2. When re-tested, HDC F174, F175 and F178 did not reduce root rot severity in the *Viola* sp. plants when applied either preventatively or curatively following Cercobin WG application. However, T34 Biocontrol at sowing before either HDC F174 or F175 significantly reduced the root area rotted from 67.2% in the untreated inoculated to 42.7%.
Objective 4: To utilise the results from work carried out under Objectives 1 to 3 to select products for application to *Choisya* sp. liners to protect against black root rot (Experiment 4; 2015)

In April 2015 *Choisya* sp. were potted up as liners at a nursery site. Drenches of HDC F178, T34 Biocontrol, Trianum-P, Prestop or Serenade ASO were applied the day after potting and repeated after five weeks at the same time as chemical drenches were applied to other plots. Cercobin WG, Signum, HDC F174, F175 and F178 were applied only preventatively and HDC F174, F175 and F178 were also re-applied 10 weeks after potting, following inoculation six weeks after potting. In August, foliar vigour was significantly better after treatment with F175 a week before inoculation than in untreated uninoculated plants. By December 2015 there was no significant difference in root rot (mean 30%) between these treatments and untreated plants.

Objective 5: To utilise the results from work carried out under Objectives 1 to 4 to select products for application to *Choisya* sp. finals to protect against black root rot (Experiment 5; 2016/17)

In April 2016 products were applied to *Choisya* sp. finals in the same nursery glasshouse as the liners that were destructively assessed in December 2015 (*Figure 2*) with protectant and curative treatments as shown in *Table 2*. Foliar vigour did not differ between treatments at any of the eight assessments. From August some *Choisya* sp. pots were yellowing and by the destructive assessment in May 2017 a number had wilted and desiccated and had fewer roots, however there were no significant differences between treatments. *Fusarium oxysporum* was isolated from a wilted plant’s roots, suggesting that this fungus, not *T. basicola*, was the cause of wilting. All treatments developed a similar level of root rot leaving on average 26% healthy root area. *T. basicola* resting spores were confirmed in dark brown patches on roots and microscopic examination showed that cells containing less-pigmented mycelial growth of the fungus spread beyond these, with the dispersal spores being released from the roots.

*Table 2.* Fungicides and biofungicide programmes examined on *Choisya* sp. finals in 2016.

<table>
<thead>
<tr>
<th>Treatment</th>
<th>At potting protectant drench or spray</th>
<th>At 5 weeks protectant drench</th>
<th>Inoculation T. basicola at 6 weeks</th>
<th>At 10 weeks curative drench or spray</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Untreated</td>
<td>Untreated</td>
<td>Y</td>
<td>Untreated</td>
</tr>
<tr>
<td>2</td>
<td>Untreated</td>
<td>Untreated</td>
<td>N</td>
<td>Untreated</td>
</tr>
<tr>
<td>3</td>
<td>Cercobin</td>
<td>Prestop</td>
<td>Y</td>
<td>Prestop</td>
</tr>
<tr>
<td>4</td>
<td>T34 Biocontrol</td>
<td>Prestop</td>
<td>Y</td>
<td>Serenade ASO</td>
</tr>
<tr>
<td>5</td>
<td>Trianum-P</td>
<td>Prestop</td>
<td>Y</td>
<td>Serenade ASO</td>
</tr>
<tr>
<td>6</td>
<td>Serenade ASO</td>
<td>Prestop</td>
<td>Y</td>
<td>Serenade ASO</td>
</tr>
<tr>
<td>7</td>
<td>Prestop</td>
<td>Prestop</td>
<td>Y</td>
<td>Serenade ASO</td>
</tr>
</tbody>
</table>
**Financial Benefits**

A review of Choisya root rots (HNS 169; Talbot & Wedgwood, 2009) listed *Fusarium* spp., *Phytophthora* spp. and *Pythium* spp. as well as *T. basicola* as common causal agents. Fungicides are thus used in preventative programmes by most growers. The cost of treatment is not great, but there are currently limited chemical options (risking resistance development) and only limited information on the efficacy of recently approved biofungicides. A Cercobin WG drench to *Viola* sp. could cost around £200 for a batch of 2500 plug trays (8 p per plug tray). With *Choisya* sp. the cost of Prestop treatment against a range of pathogens on 1000 plugs could be around £1.50 (1000 plugs selling at £280-£350), increasing to £15 per 1000 liners (1000 liners selling at £900-£1150). A Cercobin WG drench to *Choisya* sp. liners against fungal (not oomycete) root rots, could cost around 8.3 pence per 1000 pots.

Nationally, in England and Wales, it is probable that 1% of pansies are killed by black root rot, equating to an annual loss of £21,000, but this would rise to £105,000 if fungicides were not used. Around 5% overall of *Choisya* sp. and *Skimmia* sp. are probably lost to black root rot. These are the main HNS subjects affected and they represent about 2% of the container plant range and so this would equate to annual losses to black root rot in the UK from these plants alone of £346,000. Losses can however be around 20% on some nurseries in years where controls fail. Providing a range of products that can be applied at intervals during production to improve root system performance will be particularly important for hardy nursery stock where plants are sold by pot size and where greater losses arise when older plants are lost to disease.

**Action Points**

- Check plugs and liners have visibly healthy roots on delivery and before potting-on.
- Do not rely on the use of fungicides to maintain plant health as they will not be able to cure established root diseases, they only help to prevent new pathogen colonisation.
• Pay close attention to nursery hygiene such as the disinfection of trays and matting as the mycelium and resting spores (chlamydospores) can survive for several years.

• Avoid plant stress to prevent facilitating the entry of pathogens and encouraging black root rot and other rots to develop. See the HDC PO 14 review for further guidance.

• Be aware that early infection stages may cause the roots to become pale brown and typical dark brown speckling will not be seen until resting spores (chlamydospores) are formed. By the time reduced foliar vigour is seen root death could be advanced.

• Treat or destroy (do not compost) affected plants promptly otherwise endoconidia (dispersal spores of black root rot) can infect other plants and produce further sporulation within weeks.

• Seek confirmation of the cause of any plant losses so that the most appropriate control measures can be applied and avoidance sought for future batches.

• Consider preventative use of microbial products to increase resistance to a number of pathogens. Prestop (Gliocladium catenulatum) and Trianum-G (Trichoderma harzianum) are permitted as drenches to ornamentals.

• Look to select a range of chemical plant protection products with different modes of action to avoid the build-up of resistance to active ingredients and integrate with biofungicides alongside cultural controls to reduce disease susceptibility.