



Horticultural
Development
Company

Grower summary

M 48

Trichoderma green mould –
diagnostic assays for improved
disease management

Final Report 2010

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Before using all pesticides check the approval status and conditions of use.

Read the label before use: use pesticides safely.

Further information

If you would like a copy of the full report, please email the HDC office (hdc@hdc.org.uk), quoting your HDC number, alternatively contact the HDC at the address below.

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Headline

- A rapid molecular diagnostic test for *Trichoderma aggressivum* identification is now available that can permit early diagnosis of the disease and help mitigate financial losses.

Background and expected deliverables

The fungus *Trichoderma* is comprised of numerous species of which only some cause economic losses to the Mushroom Industry. At the onset of the problem in the mid 1980s, *Trichoderma* (green) compost mould and cap spotting was attributed primarily to *T. harzianum* leading to between 30-100% losses. However, several studies that investigated *Trichoderma* outbreaks in Europe and North America (Fletcher, 1986; Seaby, 1996; Muthumeenashi *et al.* 1998) showed the situation to be much more complex. There are four biotypes, some of which are present in the UK and some of which are economically damaging as summarised below:

Biotype	Current Name	Present in UK	Economically damaging
Th1	<i>Trichoderma harzianum</i>	yes	no
Th2	<i>Trichoderma aggressivum f. europaeum</i>	yes	yes
Th3	<i>Trichoderma atroviride</i>	yes	no
Th4	<i>Trichoderma aggressivum f. aggressivum</i>	no	yes

Rapid diagnosis of *T.aggressivum* cannot currently be achieved quickly so this prevents effective disease management. Recent advances in our understanding of *T.aggressivum* molecular biology (Project M 46) may permit the development of species - specific diagnostic tools.

Summary of the project and main conclusions

The project successfully developed extraction methods for *Trichoderma* DNA directly from Phase III compost. Real-time PCR primers and probe for *T.aggressivum* were developed using the translocation elongation factor gene. It was not possible to develop an assay for *T.harzianum* due to the complexity of this species taxonomy. The *T.aggressivum* assay was found to be as sensitive as plating out coupled with a standard PCR assay, but importantly,

could be achieved within one working day as opposed to the existing two stage method taking at least 8 days. The real-time PCR test can be semi-automated to help reduce time and costs and is also semi-quantitative. Levels of *Trichoderma* can be determined to at least 4500 propagules per gram of fresh weight and potentially 10 times more dilute (450 propagules/gfw; equivalent to only a few conidial heads of *Trichoderma*). The new assay has been used to successfully test several compost samples from commercial premises for *T. aggressivum*, This will be invaluable to aid the understanding of the pathogen's epidemiology in project M 50.

Financial benefits

A rapid molecular diagnostic test for *Trichoderma aggressivum* identification is now available that can permit early diagnosis of the disease and help mitigate financial losses.

Action points for growers

- Aggressive forms of *Trichoderma* compost mould (*Trichoderma aggressivum*) can cause significant yield losses (in excess of 50%) due primarily to bare areas and cap spotting.
- Early detection and rigorous hygiene measures are key to successful *Trichoderma* management.
- A semi-automated DNA extraction for Phase III compost has been developed permitting rapid diagnoses (<48h) from compost.
- The project has led to the development of the first rapid, cost effective commercial service for compost producers and growers, to screen Phase III compost directly for *T. aggressivum*.