



Grower Summary

M065

Improved understanding and control of bacterial blotch and green mould in mushroom production

Project title: Improved understanding and control of bacterial blotch and green mould in mushroom production

Project number: M065

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The results and conclusions in this report are based on an investigation conducted over 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.

[Name]

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GROWER SUMMARY

Headlines

- New blotch causing *Pseudomonas* spp. and green mould causing *Trichoderma aggressivum* f. *europaeum* have been isolated from UK farm samples
- New real time PCR assays are being developed to identify groups of blotch causing *Pseudomonas* that were not detected using previously developed assays targeting *P. tolaasii* and '*P. gingeri*'
- Irrigation of casing with a non-pathogenic pseudomonad strain showed promising control levels of blotch caused by '*P. gingeri*' and *P. costantinii*, but further tests are needed

Background

Bacterial blotch caused mainly by *Pseudomonas tolaasii*, '*P. gingeri*' and *P. costantinii*, is considered to be the most important disease currently faced by the mushroom industry in the UK and elsewhere in Europe causing losses that can frequently exceed 30% of production. Conditions that favour high yield are also favourable for disease development and transmission, and therefore there is a trade-off between maximising yield and maintaining health and quality of production. Growers would benefit from tools that allow early detection of disease and understanding the possible sources of infection. Practical control measures that can reduce or eliminate spread of pathogenic pseudomonads without having a negative impact on beneficial populations that are necessary for mushroom production, need to be developed to be used across the supply chain to reduce losses and production costs and guarantee sustainable supply.

Although the number of outbreaks of compost green mould caused by *Trichoderma aggressivum* f. *europaeum* has been reduced through the implementation of sanitation measures, this disease still occurs in some farms causing significant losses. The American form of the pathogen (*Trichoderma aggressivum* f. *aggressivum*) so far has not been detected in the UK but constitutes an additional threat to mushroom production. Early detection of *Trichoderma* species has the potential to be used to inform control strategies and also to monitor general farm hygiene.

This project follows on from project M063 and the main aims are to detect, monitor and control blotch-causing pseudomonads and *Trichoderma* species whilst retaining populations of beneficial microorganisms in mushroom cultivation. In particular the aims are:

1. Enable sensitive detection in fresh substrates of all blotch-causing *Pseudomonas* species to determine if the analysis relates to the occurrence of blotch, thereby predicting disease risk
2. Determine the relative abundance of blotch-causing pseudomonads, *Trichoderma* species and other microorganisms in mushroom cropping substrates from different sources and in response to control treatments at different stages of commercial production
3. Estimate degree of control of blotch and/or green mould achieved by irrigating with antagonists, bacteriophages and ionic solutions
4. Make diagnostic tests available and disseminate the results to the mushroom industry

Summary

Severe brown blotch, strong and mild ginger blotch and pitting observed in several UK farms was identified as *Pseudomonas tolaasii*, '*P. gingeri*' and *P. costantinii* respectively. A number of isolates still await identification. Pathogenicity was confirmed for a range of isolates in cap droplet inoculation tests and pot culture tests. Inoculation of casing with *P. costantinii* also resulted in a marked reduction in the number of healthy mushrooms harvested.

A new method to test the pathogenicity of mushrooms in small pots enclosed in plastic bags has been developed and used successfully.

Ten new TaqMan assays have been developed in the current project based on recently obtained whole genome sequences, to detect groups of pathogenic *Pseudomonas* that were not detected by previously developed real-time assays that target *P. tolaasii* and '*P. gingeri*' (project M063).

Results of TaqMan assays targeting *P. tolaasii* and *P. gingeri* (developed previously in project M063) on casing extracts corresponded with the pseudomonad isolates (*P. tolaasii* or '*P. gingeri*') that were inoculated into the casing, whereas samples inoculated with *P. costantinii* tested negative with these assays. Incubation of the casing samples with compound B increased the pseudomonad count in the extract using a plating method but did not improve the resolution of the TaqMan assay.

Inoculation of the casing with a non-pathogenic pseudomonad (isolate *P. 'reactans'* P7759) suppressed blotch caused by '*P. gingeri*' and *P. costantinii* in a small pot assay. Commercially

available pseudomonads including *Pseudomonas putida*, *P. fluorescens* and *P. chlororaphis* did not reduce the incidence of blotch.

Irrigation with ionic solutions did not consistently reduce the incidence of blotch in controlled small pot assays.

Results of sequencing of two partial genes of strains of *Trichoderma* species from a culture collection hosted at FERA and from recent farm isolates resulted in changes to the original culture designations. Cultures obtained from mushroom substrates show that *T. aggressivum* f. *europaeum* was prevalent on two farms.

Bacteriophages that target *P. tolaasii* and some strains of '*P. gingeri*' were obtained and will be further characterised.

Financial Benefits

Although it is too early to state and calculate the financial benefits of this work, the development of assays that can detect most blotch causing *Pseudomonas* and *Trichoderma* species and also the identification and testing of potential biocontrol agents can potentially lead to significant financial benefits.

Action Points

- Diagnostic tests for *Pseudomonas* causing blotch are available at Fera Science using qPCT assays that target *P. tolaasii* and '*P. gingeri*' (developed in the M063 project) and although these tests do not cover all blotch causing pseudomonads, they are still recommended for these two important groups of pathogens.
- Further work is needed to finish developing tests for diagnostics of other blotch causing pseudomonads and for green mould and to develop and test potential biocontrol agents. These results will then be disseminated to growers.