

Project title: Novel ways of managing tree crop fungal diseases: Using precision diagnostic technologies to tailor disease management strategies

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Project leader: Thomas Heaven, University of Lincoln

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Key staff: Dr Helen Cockerton
Prof Matthew Goddard

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Industry Representative: Megan McKertcher, Worldwide Fruit

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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]

[Position]

[Organisation]

Signature Date

[Name]

[Position]

[Organisation]

Signature Date

Report authorised by:

[Name]

[Position]

[Organisation]

Signature Date

[Name]

[Position]

[Organisation]

Signature Date

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GROWER SUMMARY

Headline

Collection, culture and DNA extraction methods for studying apple scab and powdery mildew genetics have been established and optimised. A high coverage genome assembly of apple powdery mildew has been generated – the first for a European isolate. Fungicide resistant strains of apple scab have been isolated.

Background

To better inform the disease management strategies of growers there is an immediate need for improved diagnostic testing for orchard diseases. Specifically, a test is needed that provides a quick diagnosis at a low cost to growers, ideally simultaneously, for a panel of diseases commonly found in orchards. This test also needs to be precise enough to identify key pathogen characteristics such as resistance to certain fungicides and virulence. One approach is a diagnosis by sequencing method which identifies genetic markers associated with resistance and virulence traits. Such a diagnostic could also be used to better track the effects of different planting decisions and agronomic practices, improving disease management over the longer term.

Commercial apple cultivation is hampered by its susceptibility to a number of fungal pathogens. The most serious among these is apple scab caused by the pathogen *Venturia inaequalis*, where production losses can be as high as 70% in heavily affected orchards (Biggs 1990; MacHardy 1996). Apart from scab the most common fungal disease found in UK apple cultivation is apple powdery mildew caused by *Podosphaera leucotricha* (Lesemann and Dunemann 2006).

Unfortunately, genomic resources for these two pathogens lag behind those available for the apple host or for other pathogenic fungi. The genetic basis for resistance to commonly used fungicides is not well understood in either scab or powdery mildew. Similarly, no virulence genes have been characterised in either species that facilitate the suppression of a host's natural immune response. Indeed, there were no genome assemblies available for apple powdery mildew prior to 2020. There has long been significant interest in developing new varieties of apple combining the fruit quality of commercial cultivars with the superior disease resistance of wild species. However, in the past the resistance of cultivars has quickly been broken in field conditions, to prevent this from happening in the future a better understanding of the pathogen is needed (Caffier et al. 2014, Caffier and Laurens 2015). The objective of this project is to generate some of the first genomic resources for the study of apple powdery

mildew. We aim to improve understanding of virulence in apple scab and powdery mildew by identifying putative virulence and avirulence genes that determine isolate host specificity as well as mutations leading to fungicide resistance. This will inform future management strategies and breeding efforts.

Summary

In the first two years of this PhD, techniques for collection, culture and DNA extraction from apple scab and powdery mildew have been refined. These methods have facilitated DNA and RNA sequencing of the apple powdery mildew genome as well as a number of other mildew species infecting Rosaceae crops. Comparison of these species genomes and transcriptomes will enable identification of key virulence and fungicide resistance factors. Additionally, strains of apple scab have been isolated for further study including fungicide resistant strains.

- Apple Scab was sampled from indicator trees known to carry resistance genes
- Apple Scab was sampled from problem orchards and confirmed to be fungicide resistant
- Fungicide resistant and susceptible strains of apple scab were crossed to generate a mapping population for the fungicide resistance trait
- Samples of powdery mildew were collected from susceptible apple, strawberry and raspberry plants
- DNA extraction and sequencing was performed on apple powdery mildew samples
- RNA extraction and sequencing was performed on strawberry powdery mildew samples
- A mapping population of apple trees were scored for susceptibility to apple powdery mildew

Financial Benefits

Apple growers are currently heavily dependent upon frequent fungicide applications to control disease in orchards. Without these products outbreaks inflict serious crop losses of up to 70%. There is increasing pressure to reduce to use of such crop protection products. This project lays the groundwork for diagnosis by sequencing of the two diseases apple powdery mildew and scab. Precision diagnosis this kind of rapid and precise diagnosis would allow the effects of management strategies to be tracked and modified, supporting the maximisation of yield whilst minimising application of fungicides.

Action Points

- There are no grower actions points at this stage of the project.