Grower Summary

CP 184

Downy Mildew & Late Blight Control

Annual report 2020

Project title:	Downy Mildew & Late Blight Control
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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.

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GROWER SUMMARY

Headline

Despite the dry season resulting in relatively few downy mildew outbreaks combined with the effects of COVID 19 regulations restricting sampling opportunities, KE and laboratory access, some progress was made with typing of a limited number of *Bremia* isolates (3), initial knowledge transfer activities, and comparisons of molecular methodologies for testing of seed lots of basil and spinach. These tests showed that:

1) simply measuring levels of DNA present in seed and washings samples could lead to overestimates of viable pathogen levels;
2) despite this, measurements of either RNA levels or of DNA by qPCR after application of propidium monoazide (PMA) both showed promise in realistically estimating viable pathogen presence;
3) steam treatments of seed lots effectively reduced pathogen DNA by 50%.

Wherever possible, tasks not completed in 2020 have been rescheduled to 2021 (e.g. program of KT and the fungicide resistance testing of *Bremia* isolates).

Background

The oomycetes are a large group of fungus-like organisms many of which have evolved to become pathogens of plants. A large and varied group of oomycete plant pathogens are spread by air-borne and/or water-splashed propagules and cause diseases primarily of the above-ground parts of plants are collectively known as the Aerial Oomycetes. Horticulturally significant pathogens within this group are the **downy mildews** (main genera in horticultural crops: *Peronospora, Hyaloperonospora, Pseudoperonospora, Plasmopara, Bremia*), stem rots, shoot diebacks and blight caused by *Phytophthora* spp. as well as shoot and leaf 'blisters' caused by *Albugo* spp.

Diseases caused by aerial oomycetes typically exhibit rapid epidemics, which if left unchecked under optimal environmental conditions have the potential to cause complete crop loss either directly by mortality, or by rendering foliar and fruit produce unmarketable. Disease control options are limited or under-utilised and currently management is heavily reliant upon the use of fungicides, often used prophylactically, as none of the available chemicals can reliably achieve curative control, and once disease is observable in crops it will often already have become established and difficult to manage. Unfortunately, the number of currently available fungicides is becoming very restricted as a result of product withdrawals and too few new introductions. The resulting reduction in the number of active ingredients being used in control programs greatly increases the risk of pathogen populations developing fungicide resistance. The use of resistant varieties, where available, is a good disease management option although their use puts huge selection pressure on oomycete pathogen populations for new races capable of overcoming host resistance. Cultural disease management methods (e.g. appropriate tilth management, removal/treatment of crop debris, manipulation of environmental conditions), often have a limited impact on disease when used alone but can greatly (even synergistically, e.g. control of {a non-oomycete with analogous epidemiology} *Botrytis* grey mould in ornamentals, O'Neill *et al*., 2002), increase the efficacy of chemicals and plant resistance in integrated management programs. Similarly, the use of rapid pathogen detection and disease simulation models can optimise the timing of fungicide applications and in some seasons reduce their number – increasing efficacy whilst reducing costs and potential environmental impacts.

The use of contaminated seeds is considered responsible for many outbreaks of downy mildews on basil and spinach, caused by *Peronospora belbahrii (Pb)* and *Peronospora effusa (Pe),* respectively. *Pe* is a seed-borne pathogen, producing heterothallic oospores in the seed coat (Kandel *et al.,* 2019) that cause systemic infection in the crop. The transmission of *Pb* on or in seed is less clear; only a single case of oospore production has been reported in basil (Cohen *et al.,* 2017) and most new disease outbreaks have been attributed to asexual aerial conidia and become evident only on relatively mature plants (Budge, Personal communication). *Pb* conidia have been observed in basil seed samples (Falach-Block *et al.,* 2019; Wood, 2021, personal communication) however, it is unlikely the propagules would remain viable for extended periods under unfavorable environmental conditions. Therefore, it is postulated that disease is propagated through mycelial infection inside the seed-coat (Jennings *et al.,* 2017).

Integrated pest and disease management (IPM) is an increasingly important and pertinent area of research for horticulture and this project aims, through the provision of reviews and best practice grower guides, to consolidate current knowledge, ensuring that measures that can be taken up are quickly disseminated and potential barriers to uptake identified. The project builds on current knowledge of several pathosystems (specifically downy mildew on lettuce, spinach and basil and to a lesser extent onion) to develop and validate the tools required for a long-term integrated approach to disease management. New tools for the genotypic analysis of *Bremia lactucae* populations, linked to phenotypic characteristics such as 'race' and fungicide sensitivity, will allow an understanding of population diversity to directly inform resistance deployment and breeding and fungicide stewardship to be greatly improved using an approach that has previously been highly successful for potato late blight (*Phytophthora infestans*, Ritchie *et al*., 2018). The other main strand of research focusses on identifying/verifying primary inoculum in spinach and basil by detection and viability-testing of seedborne infection to steer future integrated management both by improved quality screening and providing effective tools for assessing cultural controls. Here we report on the second year's progress with limited KE, assessing pathogen races and progress with assessing molecular procedures for determining levels of viable downy mildew present in contaminated seed lots.

Summary

As stated above, a combination of a low incidence of downy mildew disease and the consequences of COVID 19 restrictions severely hampered progress with this project during 2020. Despite distributing a reasonably large number of mildew sampling packs to growers and consultants, only a limited number were sent to JHI as a consequence of the low incidence of disease outbreaks. This situation was further exacerbated by disruption to postal delivery services which resulted in delays and many samples degrading en route to a condition where it was not possible to recover viable isolates. Nevertheless some progress was made especially with seed testing work and the following tasks and findings were achieved:

- Three viable isolates of *Bremia lactucae* were obtained and typed using IBEB differential varieties and all appeared to closely resemble IBEB race BI:24EU. These isolates were stored for fungicide resistance testing in 2021.
- Molecular tests have been demonstrated to be capable of detecting and quantifying small amounts basil and spinach downy mildew (DNA/RNA) both externally and inside contaminated seed lots.
- Seed lots containing low levels of downy mildew (DNA/RNA) still potentially pose a high risk for growers
- Across the seed-lots tested a greater quantity of downy mildew DNA was detected inside the seed coat compared to seed washings.
- Steam-treated basil seed lots contained approximately 50% less downy mildew DNA than untreated samples from identical lots, indicating that steam-treatment reduces the pathogen load.
- RNA levels are generally considered to be more representative of the amount of viable pathogen present than DNA levels. In both washings and inside seed RNA levels were generally ten times less than DNA levels, showing that detection of pathogen

DNA levels can lead to over-estimation of the amount of potentially viable downy mildew. Nevertheless, RNA levels increased after steam-treatment relative to untreated seed, indicating a potential response in the pathogen to steam and that RNA may remain viable even when downy mildew has been neutralised. More work is needed to clarify this situation.

- Results of the work carried out on the project have been presented in five oral presentations at grower meetings and AHDB webinars.

Financial Benefits

Aerial oomycete infections significantly reduce crop yield, with those affecting plants in propagation, in particular, able to cause total crop loss, and those in ornamentals potentially causing the crop to become unmarketable (Wedgwood, *et al.,* 2016). Timely intelligence concerning prevalent phenotypes present in downy mildew populations has potential for significant financial benefits in terms both of managing fungicide resistance and the deployment of cultivars with suitable resistance genes. In addition, the detection and interception/treatment of infected seeds is likely to have a large impact on downy mildew incidence in crops such as basil, whilst the effective use of cultural controls and decision support systems could both reduce the frequency of spray applications, improve their efficacy and reduce the pressure selecting for new pathogen genotypes with fungicide resistance and/or capable of overcoming cultivar resistances.

Action Points

Action points are still not appropriate at this stage of the project.