Using molecular tools to investigate the role of tuber-and soil-borne inoculum of *Phytophthora infestans*

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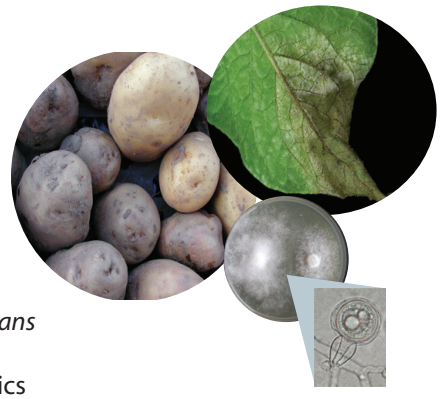


Background

- The oomycete late blight pathogen *Phytophthora infestans* infects potato foliage and tubers, reducing yield and quality of ware and seed potato crops.
- An increase in the frequency of one genotype of the A2 mating type of *Phytophthora infestans* has led to concerns that more sexual oospores may form in UK crops.
  - oospores can survive in soil for many years and germinate to initiate early epidemics
  - sexual reproduction as a result of oospore formation provides the potential for increased recombination within the pathogen population, enhancing the ability to overcome control measures.
- These changes in the UK *P. infestans* population over recent years may also influence the relative importance of tuber infections as a source of inoculum.

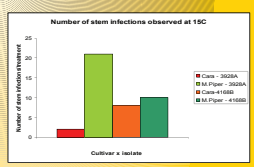
Project outline

SSR markers and a quantitative real-time PCR assay specific to *P. infestans* previously developed at SCRI have been used as tools to investigate the effect of various factors on tuber blight, stem blight and tuber/oospore derived infections. Examples of this work are described

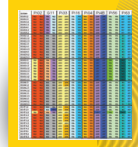
Stem blight

2 isolates of *P. infestans* (3928A = an isolate of the current dominant A2 clone '13_A2' in the UK and 4168B = a representative A1 isolate) were used to infect tubers of cultivars Cara and Maris Piper (tuber blight resistance ratings of 7 and 5 respectively).
Inoculated tubers were planted in the centre of 30cm pots and 2 uninoculated tubers were planted equidistantly from the inoculated tuber. Plants were then grown under controlled environmental conditions at 15˚C and 20˚C.
Stem base symptoms were recorded at 5 sampling dates and DNA from each lesion was analysed using SSR markers.

- All lesions tested in each pot were shown to have been caused by the isolate used to infect the central tuber.
- There was no significant effect of temperature on lesion development.
- Significantly more lesions (p=0.001)were observed on stems of cultivar M. Piper.
- Cultivars showed no difference in response to isolate 4168B but there was a significant interaction (p=0.001) between isolate 3829A and cultivar.



Oospores

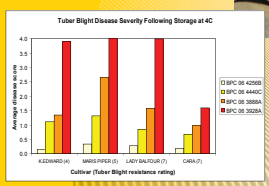


There is no evidence from SCRI *P.infestans* population studies that oospores are currently a source of inoculum for UK Late blight epidemics - relatively few clonal lineages have been observed.

Oospores derived from various crosses of parental isolates of known SSR genotype have not been shown to result in disease symptoms under field or controlled environment conditions in our experiments.
In all cases late blight symptoms could be attributed by SSR analysis to air-borne inoculum.

A test to determine levels of oospores in soil samples using real-time PCR is under development - this is necessary to quantify the effects of various factors on oospore biology

Tuber Blight



Tubers of the 4 cultivars King Edward (tuber blight resistance rating 4), Maris Piper (5), •Lady Balfour (7) and Cara (7) were inoculated with one of 4 isolates representative of the current population including the dominant clone 3928A. Tubers were incubated at 4°C for 12 weeks and the incidence and severity of tuber blight (0-4 scale) recorded.

- As expected, there was significant variation for tuber blight resistance according to resistance rating, although Lady Balfour was less resistant than expected.
- There was also significant variation for agressiveness between isolates with isolate 3829A being the most aggressive.
- Once again there was an interaction between cultivar Cara and isolate 3829A, with significantly less disease occurring compared with other cultivars.

In field experiments to examine the incidence of disease outbreaks attributable to various isolates of *P.infestans* <1% incidence of disease was shown to arise from inoculated tubers.
Current work is focusing on determining the effect of cultivar and fungicide combinations on the incidence of tuber infections (as measured by real-time PCR and visual inspection) from crops with characterised disease outbreaks

Conclusions

- SSR markers and real-time PCR are useful tools for studying the epidemiology of late blight.
- Changes in the *P. infestans* population have implications for disease management - tuber blight severity in storage caused by the current dominant A2 isolate was greater on commonly grown cultivars than that caused by other isolates.
- However, cultivar x isolate interactions have been demonstrated e.g. cultivar Cara is significantly more resistant to this isolate than other cultivars.
- Disregarding the interaction with cultivar Cara, there is no apparent effect of isolate, temperature or cultivar on transmission from infected mother tubers to neighbouring plants, resulting in stem lesions - this principle was demonstrated using characterised isolates.
- Future epidemiological studies will benefit from accurate quantification of oospores in soil - oospore derived epidemics have not been described in the UK to date.