Disease management in oilseed rape: insights into the *Brassica napus*-*Pyrenopeziza brassicae* pathosystem

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Introduction

Light leaf spot, caused by the fungal pathogen *Pyrenopeziza brassicae*, is the most damaging disease problem in oilseed rape (*Brassica napus*) in the United Kingdom. According to recent survey data, the severity of epidemics has increased progressively across the UK, with yield losses of up to £160M per annum in England (www.cropmonitor.co.uk) and more severe epidemics in Scotland. Light leaf spot is a polycyclic disease, with primary inoculum consisting of airborne ascospores produced on diseased debris from the previous cropping season (Gilles *et al*. 2001). Splash-dispersed conidia produced on diseased leaves are the main component of the secondary inoculum (Evans *et al*., 2003). Currently, the management of light leaf spot relies on fungicide applications, recommendations for resistant cultivars and cultural practices (AHDB, 2021). However, considering the frequent occurrence of light leaf spot epidemics in recent years, it appears that current control strategies are not sufficient to achieve successful control of light leaf spot. Even though the occurrence of severe epidemics of light leaf spot has been reported in the UK since 1970s, the knowledge about this pathosystem, especially in relation to the operation of host resistance, is very limited compared to that for other important pathosystems of oilseed rape such as phoma stem canker (*Leptosphaeria maculans*) (Boys *et al*., 2007; Karandeni Dewage *et al*., 2018). Therefore, this study focused on investigating interactions between *P. brassicae* and *B. napus* using single-spore isolates to provide better understanding of the different sources of resistance against *P. brassicae*.

Materials and methods

Single-spore isolates of *P. brassicae* were obtained from diseased leaf samples collected from oilseed rape crops and sub-cultured onto malt extract agar (MEA) plates. Pathogen inoculum was prepared from conidia produced by sub-cultured isolates using sterilised distilled water. The inoculum concentration was adjusted to 10^5 spores/ml and it was stored at -20ºC until needed. Oilseed rape genotypes that included commercial cultivars and breeding lines were selected, based on resistance ratings from the AHDB Recommended List (RL) trials and to represent diverse genetic backgrounds. Selected genotypes were grown under glasshouse conditions until they reached growth stage 1,4-1,5 (four-five leaves unfolded) and spray-inoculated with conidial suspensions prepared from single-spore isolates of *P. brassicae*. After inoculation, regular observations were made to identify the timing and appearance of different resistant/susceptible phenotypes. Final disease assessment was made at 29 days post inoculation (dpi) on plants that were destructively harvested at the stem base at 24 dpi and

incubated at 4ºC for five days in polyethylene bags. Light leaf spot severity was measured as % leaf area covered with *P. brassicae* sporulation and compared between different treatments to identify resistant/susceptible interactions.

Results and Discussion

Infected plants showed different resistant/susceptible phenotypes associated with *P. brassicae*. This included leaf deformations, necrosis and different numbers of acervuli (asexual sporulating structures of *P. brassicae*) with or without the presence of visible lesions. Leaf deformations appeared at *c.*7 dpi, indicating possible association with early stages of *P. brassicae* colonisation. Symptoms included leaf curling, leaf distortions and petiole elongations and could be seen to some extent in all the cultivars/lines tested in the experiments. Some of the cultivars/lines appeared to have necrotic responses to *P. brassicae* infection. Of these cultivars/lines, the majority had reduced numbers of *P. brassicae* acervuli, suggesting an association between these two phenotypes. Considering the light leaf spot severity measured as % leaf area with *P. brassicae* sporulation, there were significant differences ($P \leq 0.05$) between cultivars/lines and between isolates and there were significant cultivar/line-isolate interactions. Some of the pre-breeding lines showed resistance against most of the isolates tested. These lines may provide sources of resistance for oilseed rape breeding programmes. There has been little work on specific host-pathogen interactions regarding light leaf spot disease, and identification of different types of resistance available in commercial oilseed rape cultivars is important for effective deployment of cultivar resistance. This study shows the potential for studying specific cultivar-by-isolate interactions in the *B. napus* – *P. brassicae* pathosystem using single-spore isolates of *P. brassicae*. It is anticipated that this type of experimentation will help to identify different sources of resistance that can be exploited in oilseed rape breeding programmes.

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