# Development of phoma stem canker (*Leptosphaeria maculans*) and light leaf spot (*Pyrenopeziza brassicae*) on current and historical oilseed rape cultivars in 2003/04, 2004/05 and 2005/06 UK growing seasons

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**Abstract**

Winter oilseed rape (sown August, harvested July) field experiments at Rothamsted in successive growing seasons (2003/04, 2004/05 and 2005/06) were monitored for phoma leaf spot and phoma stem canker (*Leptosphaeria maculans*) and light leaf spot (*Pyrenopeziza brassicae*). Each experiment included 42 current, historical or exotic *Brassica napus* cultivars/breeding lines. Weekly assessments examined leaf, stem and pod disease as % plants affected and % area affected. Significant differences were observed between cultivars for both diseases, suggesting that the cultivars ranged from susceptible (severely diseased) to resistant (little disease) to the pathogens. Many of the cultivars that were susceptible or resistant in one season gave similar responses in the other seasons. However, material gave a range of responses to the two pathogens; for example, cv. Aviso was very resistant to *L. maculans* in all seasons whilst Bronowski was the most susceptible cultivar in all seasons. For *Pyrenopeziza brassicae*, cv. Tapidor was consistently susceptible whilst the line PR45W05 was consistently resistant, when % leaf area affected was assessed. The results are discussed in relation to current knowledge of the reported oilseed rape resistance gene mediated response to *L. maculans* and the suggested oilseed rape resistance mechanism to *P. brassicae*.

**Key words:** Brassica napus, Phoma lingam, Cylindrosporium concentricum, disease assessment, blackleg

## Introduction

The most economically important diseases of oilseed rape (*Brassica napus,* canola, colza, rapeseed, Raps) in the UK are phoma stem canker (blackleg; *Leptosphaeria maculans*, anamorph *Phoma lingam*) and light leaf spot (*Pyrenopeziza brassicae*, anamorph *Cylindrosporium concentricum*), causing an average annual loss, in 2000-2002, of €56M and €28M respectively (Fitt *et al.*, 2006). Phoma stem canker is a monocyclic disease, for which the primary inoculum is ascospores released from the debris of the previous season's crop. Characteristic phoma leaf spots are the first sign of disease. The pathogen then grows asymptomatically down the petiole to the plant stem where it is able to invade and kill host cell tissue, resulting in the formation of a damaging canker in the spring/summer (West *et al.*, 2001). Light leaf spot is polycyclic disease that affects leaves, stems, flowers and pods during the growing season (Gilles *et al.*, 2000).

There are two types of resistance against *L. maculans*, namely qualitative and quantitative resistance. Qualitative resistance (monogenic) is race specific whilst quantitative resistance (polygenic; partial) is non-race specific (Pilet *et al.*, 2001; Delourme *et al.*, 2006). Qualitative resistance, involving single major resistance genes (*Rlm* genes), is expressed in the leaves and cotyledons of *B. napus* (Delourme *et al.*, 2006). Race specific interactions between the avirulence alleles (*AvrLm*) of *L. maculans* and *B. napus Rlm* genes result in a resistance response (Delourme *et al.*, 2006; Rimmer, 2006). To date, nine *AvrLm* loci have been identified (Balesdent *et al.*, 2006; Delourme *et al.*, 2006; Rimmer, 2006). The mechanism of resistance to *P. brassicae* is not fully understood. A range of current and historical winter oilseed rape cultivars were used, in the experiments, to investigate resistance responses to field populations of *L. maculans* and *P. brassicae.*

## Materials and methods

Field experiments were done at Rothamsted Research, Hertfordshire, UK in three successive growing seasons (2003/04, 2004/05 and 2005/06). In these experiments, 42 current and historical winter oilseed rape cultivars were grown in a randomised block design with three replicates. The 2003/04, 2004/05 and 2005/06 experiments were hand sown at a sowing density of 100 seeds/m$^2$ on 11/12 September 2003, 3 September 2004 and 15 September 2005, respectively. Each cultivar/line had a border of cv. Shannon, a susceptible cultivar, to decrease edge effects and interplot interference. The field experiments received no fungicide application. The plots were inoculated indirectly with a bale of upper stem and pod debris from the previous season, to increase the severity of the light leaf spot epidemic. Weekly assessments were done once symptoms were observed. Assessments were done by randomly selecting 10 plants per replicate of each cultivar/line (30 plants per cultivar in total). The number of plants out of 10 with phoma leaf spotting and percentage leaf area affected with phoma leaf spot were recorded. The number of plants out of 10 with light leaf spot and percentage leaf area affected with *P. brassicae* were also recorded. Assessment of stem and pod disease began at the first sign of disease and continued until harvest. At harvest 10 whole plants per cultivar line were removed. Plants were assessed for phoma stem canker by cutting through the base of the stem at the root collar and scoring any canker on a 0-4 scale (0 = no canker; 4 = 100% stem girdled and discoloured) (Zhou *et al.*, 1999).

## Results

In all three seasons there was a range in severities of both phoma stem canker and light leaf spot. Incidence of phoma leaf spotting increased untill December in 2003/04 and 2004/05 and January in 2005/06 (data not shown). There was then a rapid decrease due to senescence of early leaves. In the spring/summer a large range in stem canker severity between cultivars was observed (Fig. 1). In all seasons cv. Twister and, in the 2003/04 and 2004/05 seasons, cv. Aviso were amongst the cultivars which developed least phoma leaf spotting and phoma stem canker. Cultivar Bronowski was the most susceptible in all three seasons. In 2004/05, cv. Hyola60 initially had little phoma leaf spotting; however, incidence of the disease rapidly increased during the stem disease stage. Cultivar Pollen had low leaf and stem disease in all three seasons.

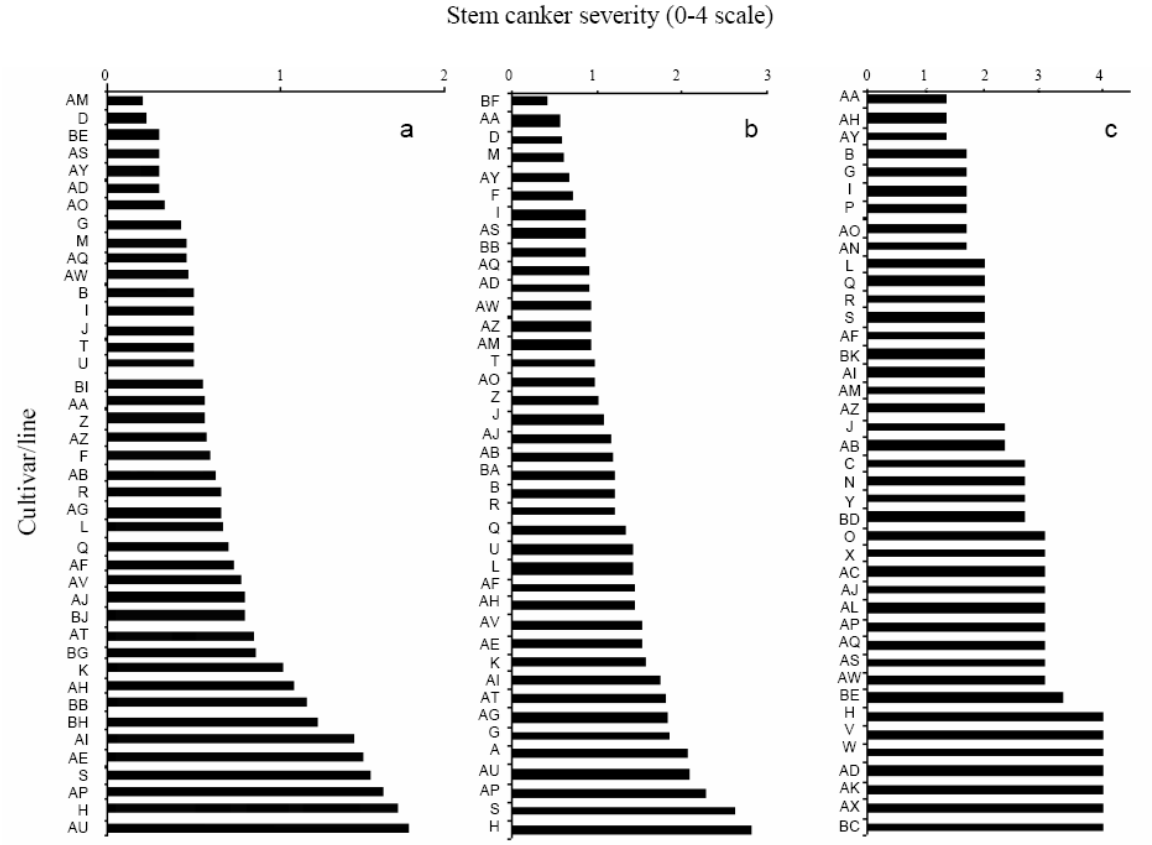


Figure 1. Stem canker severity score for oilseed rape plants affected with phoma stem canker (*L. maculans*) for a range of current or historical oilseed rape cultivars or breeding material at Rothamsted, assessed pre-harvest at the end of the (a) 2003/04 (S.E.D. = 0.39, d.f. = 75), (b) 2004/05 (S.E.D. = 0.42, d.f. 67) and (c) 2005/06 (S.E.D. = 0.34, d.f. 80) growing seasons. Cultivars Apache (A), Apex (B), Apex 93 5×Ginyou 3 DH (C), Aviso (D), Bienvenu (E), Bosman (F), Bristol (G), Bronowski (H), Canary (I), Canberra (J), Capitol (K), Cobra (L), Columbus (M), Columbus×Nickel DH (N), Comet (O), Columbus (M), Columbus×Nickel DH (N), Comet (O), Darmor (P), Escort (R), Eurol (S), Express (T), Falcon (U), Fido (V), Global (W), Hanna (X), Hansen×Gaspard DH (Y), Hyola 60 (Z), Jet Neuf (AA), Kana (AB), Karat (AC), Liho (AD), Lipton (AE), Madrigal (AF), Major (AG), Marita (AH), Mikado (AI), Mohican (AJ), Ningyou7 (AK), Nugget (AL), Pollen (AM), Rocket (PTS)×Lizard DH (AN), PR45W05 (AO), Quinta (AP), Recital (AQ), Shannon (AS), Surpass200 (AT), Synergy (AU), Talent (AV), Tapidor (AW), Tower (AX), Twister (AY), Victor (AZ), Vivol (BA), VivoP (BB), Wild Accession (BC), Winner (BD), Yudal (BE), Zenith (BF), Rafal (BG), Matador (BH), Norin (BI), Primor (BJ), Madrigal×Recital (BK).

Light leaf spot symptoms were first observed in February in 2003/04, mid December in 2004/05 and in March in 2005/06. A range of disease severities was observed (Fig. 2). Some material gave a similar response in all seasons; for example PR4SW05, Escort and Mohican were consistently resistant whilst Tapidor, Aviso and Bronowski were consistently susceptible. However, some material gave different results depending on the season; for example Surpass, which gave a resistant response in the 2003/04 season gave a susceptible response in the 2004/05 season. In contrast cv. Hyola 60 gave a susceptible response in the 2002/03 seasons and a resistant response in the 2003/04 season.

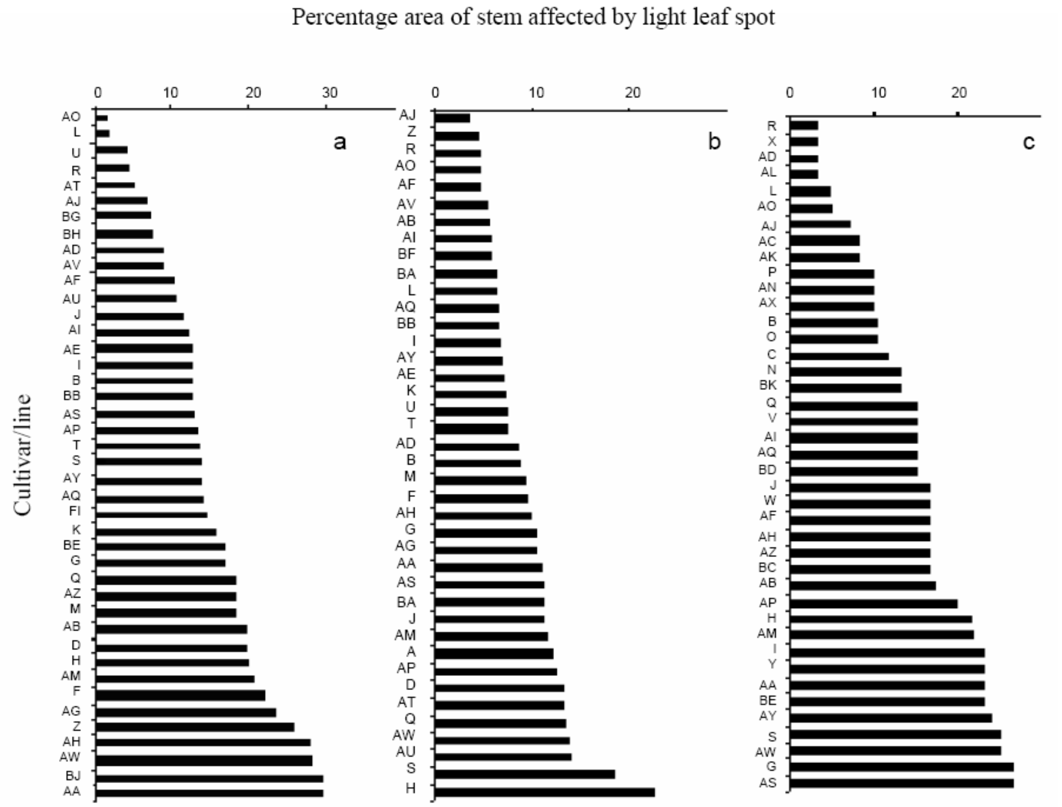


Figure 2. Percentage of oilseed rape stem area affected with light leaf spot (*P. brassicae*) for a range of current or historic oilseed rape cultivars at Rothamsted during the (a) 2003/04 (S.E.D. = 5.4, d.f. = 70) and (b) 2004/05 (S.E.D. = 6.7, d.f. = 67) and (c) 2005/06 (S.E.D. = 8.4, d.f. = 80) growing seasons. Cultivars Apache (A), Apex (B), Apex 93 5×Ginyou 3 DH (C), Aviso (D), Bienvenu (E), Bosman (F), Bristol (G), Bronowski (H), Canary (I), Canberra (J), Capitol (K), Cobra (L), Columbus (M), Columbus×Nickel DH (N), Comet (O), Columbus (M), Columbus×Nickel DH (N), Comet (O), Darmor (P), Escort (R), Eurol (S), Express (T), Falcon (U), Fido (V), Global (W), Hanna (X), Hansen×Gaspard DH (Y), Hyola 60 (Z), Jet Neuf (AA), Kana (AB), Karat (AC), Liho (AD), Lipton (AE), Madrigal (AF), Major (AG), Marita (AH), Mikado (AI), Mohican (AJ), Ningyou7 (AK), Nugget (AL), Pollen (AM), Rocket (PTS)×Lizard DH (AN), PR45W05 (AO), Quinta (AP), Recital (AQ), Shannon (AS), Surpass200 (AT), Synergy (AU), Talent (AV), Tapidor (AW), Tower (AX), Twister (AY), Victor (AZ), Vivol (BA), VivoP (BB), Wild Accession (BC), Winner (BD), Yudal (BE), Zenith (BF), Rafal (BG), Matador (BH), Norin (BI), Primor (BJ), Madrigal×Recital (BK).

## Discussion

Significant differences between cultivars in the incidence and severity of phoma leaf spotting were observed in all three seasons. Cultivars Aviso and Twister were consistently the most resistant and cultivar Bronowski the most susceptible in all three seasons. Differences in the incidence and severity of phoma leaf spotting between cultivars may have been due to differences in the *Rlm* genes possessed by the different cultivars. Qualitative resistance prevents infections through a gene-for-gene interaction between *B. napus Rlm* genes and *L. maculans AvrLm* alleles (Delourme *et al.*, 2006). Therefore, variation in *Rlm* genes between cultivars could account for differences in the incidence and severity of leaf spotting. Similarities in the resistance/susceptibility of particular cultivars over the seasons of the experiment suggest that the pathogen population did not change greatly from one season to the next. This is not surprising as the areas of plants grown in the experiments were small and successive experiments were done in fields separated by distances of up to 1 km.

The results suggest that the severity of light leaf spot epidemics is dependent upon environmental factors. There was a lack of agreement in the results of the three seasons with respect to the percentage stem area affected by light leaf spot. This indicates that disease severity is influenced by environmental factors rather than genetic control. The initial development of light leaf spot is weather dependent (Cheath & Hill, 1985; Gilles *et al.,* 2000). Early onset of light leaf spot symptoms indicates a severe epidemic. The earliest onset of symptoms, mid December, was in the 2004/05 season when October rainfall had been high (146mm). In the 2003/04 season October rainfall was also high (127.2mm), light leaf spot symptoms were observed in February. In the 2005/06 season October rainfall was low (88.4mm) and the onset of light leaf spot symptoms was later (March). This indicates that the severity of light leaf spot epidemics differs between seasons depending upon environmental conditions.

## Conclusions

Significant differences in phoma stem canker and light leaf spot were observed between a range of current and historical cultivars. The results of these experiments provide evidence for the gene-for-gene interactions between *B. napus* and *Leptosphaeria maculans*. Environmental rather than genetic factors may influence the severity of light leaf spot epidemics. The use of a more natural inoculum would provide a better test of light leaf spot resistance response. This could be achieved by doing future experiments in an area, such as Scotland, where light leaf spot is the major *B. napus* disease.

## Acknowledgements

We thank Defra (UK Department for Environment Food and Rural Affairs), ProColza, the Home-Grown Cereals Authority, The Arable Group, Chadacre Agricultural Trust and the Morley Agricultural Foundation for funding this work. Rothamsted Research receives funding from the UK Biotechnology and Biological Sciences Research Council.

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