SESSION 7C

OILSEEDS – IMPROVING MANAGEMENT OF PESTS AND DISEASES

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Papers:	7C-1 to 7C-4

A review of pest and disease problems in winter oilseed rape in England and Wales

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ABSTRACT

The status diseases and autumn pests in winter oilseed rape during 1997-2001 was reviewed using survey data from about ninety-five crops each year. Disease assessments were carried out at mid-leaf production (autumn), stem extension (spring) and pod ripening (summer). Pest and virus assessments were carried out in the autumn and spring. There have been large variations in the occurrence of pests and diseases between years and between regions despite widespread use of pesticides. Phoma canker has become the dominant disease of oilseed rape although light leaf spot remains a significant risk particularly in the north and west. There have been few problems with sclerotinia although many crops still received sprays at flowering. Trends towards use of cultivars with poor resistance to phoma and light leaf spot indicate future risks of disease epidemics. Data showed that control strategies for both pests and diseases have either been inadequate or unnecessary in many cases. Improved guidance through decision support systems should improve targeting and cost-effectiveness of inputs in the future.

INTRODUCTION

The DEFRA Winter Oilseed Rape Survey in England and Wales has been carried out annually since 1986 (Turner *et al.*, 2002). Data from these surveys have been used to quantify changes in pests and diseases and the impacts of crop protection treatments and agronomic practices on their control. The long-term national survey of winter oilseed rape diseases has highlighted major changes in fungicide control practices as a result of fluctuating disease risk, improved fungicide chemistry, economic pressures and advances in the understanding of disease epidemiology and control (Turner *et al.*, 2000). This paper reviews changes in status of the key diseases and autumn pests affecting winter oilseed rape during the last five years in relation to variations seen since the survey began.

MATERIALS AND METHODS

In each year from 1997 to 2001, approximately 95 crops were selected for pest and disease assessment from farms throughout England and Wales, the number of farms selected being in proportion to the area of rape grown in each county. The crops were sampled on three occasions: in the autumn at mid-leaf production (early December), in the following spring at early stem extension (March) and again in the summer at pod ripening (July). On each occasion, 25 plants were sampled randomly and the severity of diseases assessed. The autumn and spring samples were also assessed for cabbage stem flea beetle (*Psylliodes chrysocephala*), aphids (e.g. *Myzus persicae, Brevicoryne brassicae*), rape winter stem weevil (*Ceutorhynchus picitarsis*) and slug (*Arion* spp.) damage. Sub-samples of 10 plants were taken at random from both the autumn and spring samples and assayed for the presence of *Beet western yellows virus* (BWYV), *Cauliflower mosaic virus* (CaMV) and *Turnip mosaic virus* (TuMV) using an enzyme-linked immunosorbent assay technique. Agronomic details such as sowing date, cultivar and pesticide use were also recorded for each crop surveyed. The data collected were entered onto an INFORMIX relational database at CSL for subsequent analysis. Summary information, interactive searches, risk maps and annual reports from this project can be accessed via the web at: www.crop-disease-surveys.com.

Data for the period 1990 to 1999 have been used to generate regional maps illustrating the prevalence of oilseed rape diseases over the last decade. The survey data were interpolated using a kriging technique to produce a continuous surface of disease incidence. In order to restrict the maps to only those locations where winter oilseed rape was grown, the disease incidence data were plotted onto a grid mask of the winter oilseed rape cropping area, generated from data supplied by the Edinburgh Data Library. The darker areas on the map indicate high incidence of disease. Data for the same period have also been used to generate 10-year means for pest and disease incidence, which have been used for comparison with data for the last five years.

RESULTS

Disease incidence

Downy mildew (*Peronospora parasitica*) and phoma leaf spot (*Leptosphaeria maculans*) have consistently been the most prevalent diseases in the autumn, with phoma becoming predominant in 2000 and 2001 (Table 1). The incidence of light leaf spot (*Pyrenopeziza brassicae*) and phoma in 2001 were the highest recorded during the five-year period. While incidence of downy mildew peaked in 1996/97, levels were below the ten-year mean in the autumn of the two most recent surveys.

Table 1. Incidence of diseases in autumn (% plants affected)

	96/97	97/98	98/99	99/00	00/01	Peak (year) (1992 onwards)	Mean (1992-01)
Phoma leaf spot	21.2	29.2	26.8	30.8	51.4	60.1 (94/95)	30.4
Light leaf spot	0.3	0	0.3	0.4	0.6	1.7 (94/95)	0.5
Dark leaf spot	3.5	2.9	2.9	1.0	2.7	6.6 (93/94)	3.4
Downy mildew	51.9	44.8	39.4	20.1	24.9	51.9 (96/97)	31.6

In spring 2001 incidences of three of the four main diseases surpassed the ten-year mean, with phoma reaching the highest levels recorded since the survey began (Table 2). Levels of downy mildew (*Erysiphe cruciferarum*) exceeded the ten-year mean in all years between 1997 and 2001, peaking in 1999 with over 40% of plants affected. Light leaf spot levels have been increasing over the last three years but were well below the peak in 1994 when almost 50% plants were affected. In contrast, highest levels of dark leaf spot (*Alternaria brassicae, A. brassicicola*) during the last five years occurred in 1998.

Table 2. Incidence of diseases in spring (% plants affected)

	1997	1998	1999	2000	2001	Peak (year) (1992 onwards)	Mean (1992-01)
Phoma leaf spot	38.5	34.3	37.5	31.7	51.7	51.7 (2001)	36.1
Light leaf spot	7.3	3.5	7.8	11.9	18.0	48.8 (1995)	16.5
Dark leaf spot	0.5	4.7	0.2	0.8	1.8	5.6 (1994)	2.1
Downy mildew	40.1	32.5	40.7	31.1	33.6	40.7 (1999)	29.2

Levels of the major pod diseases in 2001 were all below the ten-year mean. Levels of dark pod spot (*Alternaria* spp.) were high for four consecutive years between 1997 and 2000 (Table 3). The highest incidence in 1998 was the highest recorded since the survey began. A mean of 2% of the pod area was affected, also the highest severity recorded since the survey began. Levels of downy mildew were highest in 2000. The most common stem disease was phoma canker (*L. maculans*), which affected 97% of crops in 2000 (the highest since the survey began), while the incidence of 57% plants affected was the highest level since 1993. Light leaf spot was the second most common stem disease, and affected 27% of stems in 2001, less than half the level recorded in the epidemic of 1994. Incidence of stem rot (*Sclerotinia sclerotiorum*) was highest in 2000. Incidence of powdery mildew was exceptionally high in 1997 compared to other years and levels of disease have since been lower by an order of magnitude.

Table 3. Incidence of diseases at pod ripening (% plants/stems affected)

	1997	1998	1999	2000	2001	Peak (year) (1992 onwards)	Mean (1992-01)
% plants							
Phoma pod spot	3.6	0.6	0.2	2.0	0.1	3.6 (1997)	0.8
Light leaf spot	13.4	7.8	7.5	12.3	8.0	38.7 (1994)	13.2
Dark pod spot	33.3	62.6	48.5	31.5	6.1	62.6 (1998)	24.0
Downy mildew	8.6	11.9	5.9	16.4	3.4	16.4 (2000)	5.9
% stems							
Light leaf spot	16.7	8.8	20.2	21.8	27.1	57.1 (1994)	26.5
Phoma canker	26.8	27.7	40.7	56.8	54.1	58.4 (1993)	40.1
Powdery mildew	19.2	1.2	1.5	1.4	2.1	19.2 (1997)	3.6
Sclerotinia	1.0	1.7	1.7	4.1	1.1	5.4 (2000)	1.8

Agronomic practice

There has been a decreasing trend in the use of cultivars with high levels of resistance to phoma and a major switch to cultivars considered susceptible to light leaf spot during the last five years (Figure 1). In 2000, 43% of cultivars grown had a disease resistance rating of 5 or less (indicating susceptibility) for light leaf spot and 65% of cultivars were considered susceptible to stem canker.

Use of fungicides on winter oilseed rape increased to over 90% of crops treated in 1998 and has since remained relatively unchanged over the last four years although there is some indication of a decreasing trend (Figure 1). The major trend has been the increase in the use of sprays applied in the autumn for phoma leaf spot and light leaf spot control. Between 1997 and 2000, there were consistent year-on-year increases in use of autumn sprays and a concurrent decrease in use of sprays applied in the spring and at flowering. Decreases in sprays in autumn 2000 were related to poor weather conditions. The decrease in use of sprays applied in the spring and summer of 1999 was linked to farmers reducing costs in response to a fall in the price of oilseed rape to £100/tonne. Use of sprays applied in spring and at flowering (mainly for sclerotinia control) increased in 2001, reversing a two-year decline. Post-flowering sprays, principally for control of dark pod spot, were used at low frequency.

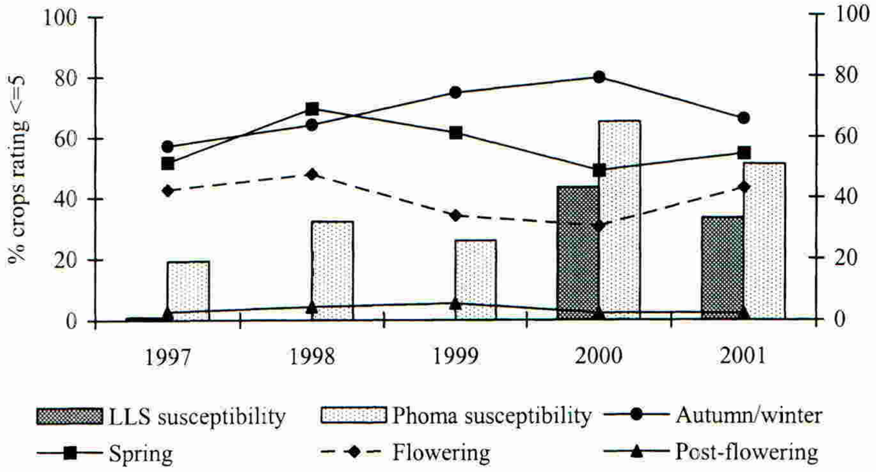

Figure 1. Use of susceptible cultivars and fungicide sprays

Incidence of viruses

The incidence of BWYV in the autumn during the five-year period was generally low with the highest incidence occurring in 1998/99 (Figure 2). Levels of virus have decreased considerably since monitoring of BWYV started in 1989/90 when a peak of 53% of plants was recorded. The prevalence of BWYV in the spring of 1998 (63% crops affected) was the highest recorded since 1991. The incidence of BWYV within crops (26% plants affected), although well below the peak level recorded in 1990, was the highest since 1992. Increases in virus incidence over-winter ranged from two to four-fold. Peaks in incidence of CaMV and TuMV occurred in the early 1990s and have since decreased markedly, not exceeding a mean of 1% of plants affected in the last five years. Consistent decreases in the prevalence of virus diseases have occurred since 1997/98, despite considerable decreases in the use of autumn insecticides during the same period.

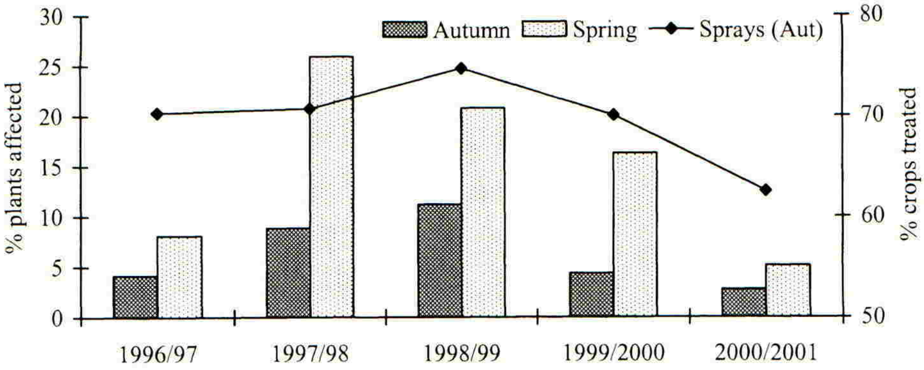

Figure 2. Incidence of BWYV and use of autumn insecticides

Incidence of pests

A mean of 0.4 cabbage stem flea beetle larvae per plant was found in the autumn 2000/01 samples, lower than in 1999/00 and breaking the trend towards increasing incidence recorded since 1997/98 (Figure 3). Only two of the total number of fields monitored during the last five years exceeded the current treatment threshold of five larvae per plant or 50% leaves scarred (Walters *et al.*, 2001). The mean number of cabbage stem flea beetle larvae in samples taken in spring has been lower than in the previous autumn in four out of the last five years. Use of autumn insecticides reached 75% of crops treated in 1998/99, but use has since declined to less than 65% crops treated in 2000/01.

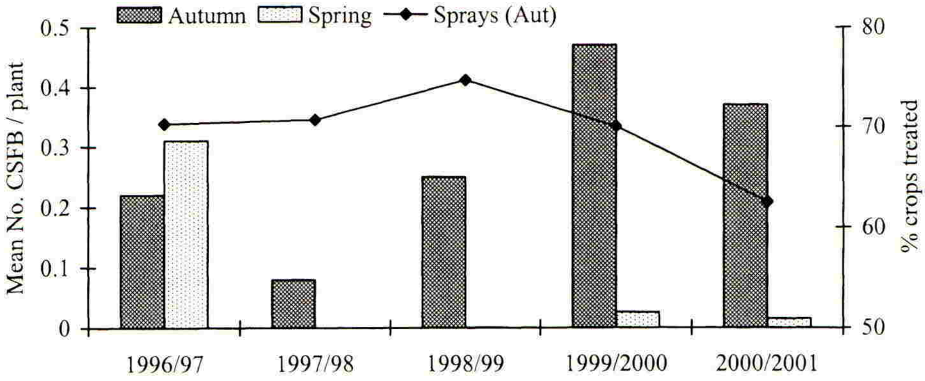

Figure 3. Incidence of cabbage stem flea beetle (CSFB) and autumn insecticide use

Regional trends in pest and disease risk

Mapping of incidence of the four main diseases of oilseed rape over the period 1990-1999 clearly illustrates the differing regional risk from individual diseases (Figures 4-7).

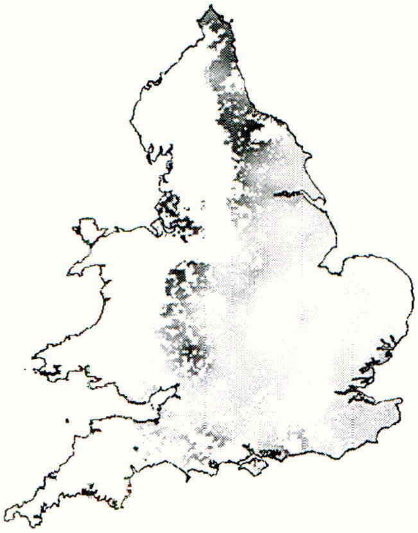

Figure 4. Incidence of light leaf spot in spring 1990 -1999

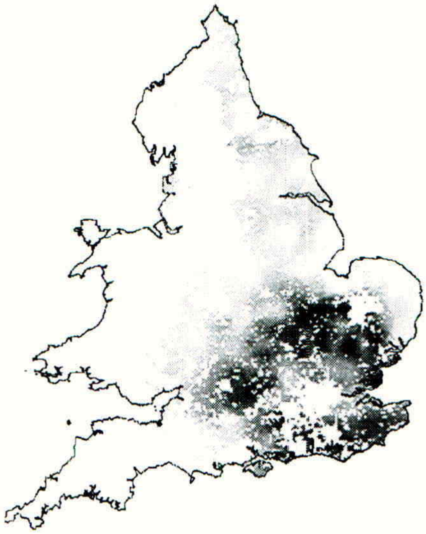

Figure 5. Incidence of phoma canker in summer 1990 - 1999

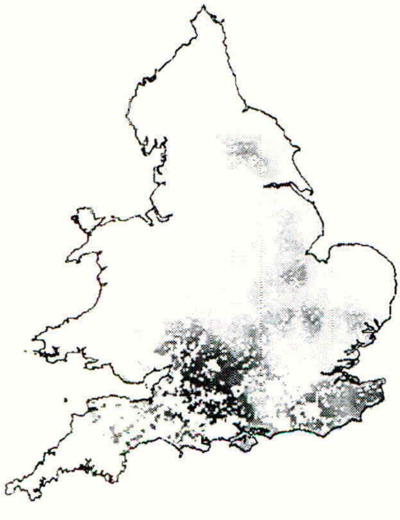

Figure 6. Incidence of dark pod spot in summer 1990 - 1999

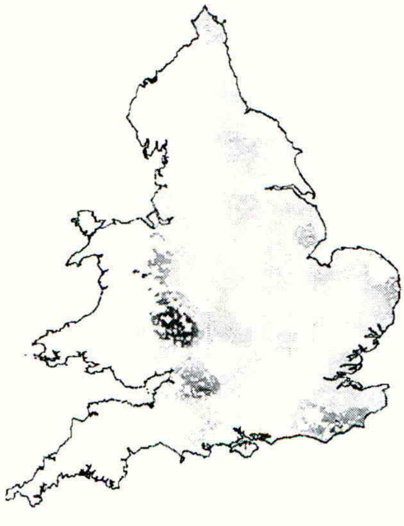

Figure 7. Incidence of sclerotinia stem rot in summer 1990-1999

Regional trends in disease incidence and severity were similar over the years 1997 to 2001, and were consistent with overall trends recorded since 1986. Levels of light leaf spot in spring were consistently highest in the north and west of England and lowest in the east whereas risks of phoma canker, now the most common stem disease of winter oilseed rape, were highest in the eastern and south-eastern counties of England. Over the last decade, levels of dark pod spot have tended to be highest in the south and west of England, largely reflecting rainfall patterns. Risks of sclerotinia have been generally low and localised, but when conditions have been conducive for disease development, risks were highest in the Herefordshire area. Use of similar techniques to examine the regional incidence of viruses and cabbage stem flea beetle has shown no obvious regional influences.

DISCUSSION

Disease levels have fluctuated considerably during the years 1997 to 2001 with incidence of some diseases reaching unprecedented levels during this period. Overall yield losses increased steadily between 1997 and 2000 reaching a peak value of over £75M in 2000 despite fungicides having been used to control disease (Turner *et al.*, 2002). Seasonal weather conditions are the likely cause of such exceptional disease levels, which have occurred despite the widespread use of pesticides and cultural control practices. Data from the UK meteorological office show that the average rainfall in 1997 was equal to the 30-year mean, and that rainfall levels in every year since have greatly exceeded this figure. This was particularly so in 2000 when total rainfall was 6% higher than average. Similarly, average temperatures for central England have exceeded the 30-year mean in nine out the last ten years. Whilst these weather conditions may be a transient feature, the possibility of climate change must be considered. Data show that if these conditions persist there will be a considerable impact on levels of pest and disease activity on an annual basis.

The levels of disease activity also indicate that the potential to control these diseases using chemical control methods is still not being fully exploited. Use of fungicides has stabilised since 1998 at just over 90% of crops treated each year with a total cost of in excess of £7M in 2001. Data show that growers are responding to disease pressure and advisor recommendations e.g. increases in use of fungicides in autumn (Turner *et al.*, 2000) but that timing and dose are not always optimal. Phoma canker is now the major threat to the industry. Sprays for sclerotinia were used widely with 43% of crops receiving a spray despite there being few serious problems since 1991. Despite levels of pest infestation being well below threshold, use of insecticides has also remained high during the survey period with up to 80% of crops treated each year. Autumn spraying additionally appears to have little effect within-year on subsequent BWYV levels. There is, therefore, major potential for rationalisation of pesticide use with the consequent environmental and economic benefits. The need for more reliable forecasts to aid the targeting of sprays for both pests and diseases is clearly highlighted; a requirement which is currently being addressed in the SAPPIO LINK-funded PASSWORD project. The benefits of cultivar resistance for disease control are also not being fully exploited. Given the current threat from diseases and the need to minimise inputs, there is clearly significant potential for growers to minimise disease problems by utilising cultivars that have a higher resistance rating to principal threats from stem canker and light leaf spot.

Mapping techniques have demonstrated clear regional risks for incidence and severity of oilseed rape diseases. There is undoubtedly a requirement for more accurate assessment of local disease risk in order to guide targeting of fungicide applications in order to improve gross margins, particularly given the increasing economic and environmental pressures on farming. Use of the survey maps should assist in raising awareness of risks of disease in key areas of the country and, perhaps more importantly, reduce unnecessary spraying in areas of very low risk, particularly when combined with the use of resistant cultivars. Furthermore, a forecasting scheme based on these data has been developed to predict light leaf spot levels (Fitt *et al.*, 1998) and a model to predict phoma canker is now being developed through the PASSWORD project. The lack of regional trends in virus incidence was surprising as warmer areas of the country might have been expected to be more severely affected due to better survival/reproduction of the aphid vector. However, this lack of relationship does suggest that other factors such as seasonal weather conditions and sowing date (in the case of BWYV) were of greater influence than, or at variance with, the regional effect.

The influence of agronomic factors on pest and disease incidence and severity, such as date of sowing and spray timing, can be clearly identified and there are indications of major risks for the future, particularly from phoma. Diseases are a continuing threat to the production of oilseed rape but effective control can be achieved. Control strategies will need to be reassessed to encompass a more targeted and integrated approach exploiting cultivar resistance, cultural practice and forecasting alongside chemical control. This will result in additional economic and environmental benefits and aid the move towards sustainable crop production. The current weather patterns and the recorded use of routine pesticide applications only serve to highlight the need for more reliable forecasts and risk assessments in order to target treatments more effectively while minimising yield loss. Development of such forecasting and decision support systems should remain a priority, with these surveys providing a vital resource in their development and validation.

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New perspectives on the epidemiology and management of phoma stem canker of winter oilseed rape in England

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ABSTRACT

The epidemiology of phoma stem canker (*Leptosphaeria maculans*) on winter oilseed rape will be discussed in relation to results of experiments comparing fungicide regimes in southern England. Factors affecting the severity of epidemics in England include: 1. The population of *L. maculans*; A-group and B-group *L. maculans* differ in ascospore germination, penetration of leaves, leaf spot symptoms and position and severity of stem cankers/lesions. 2. Maturation of ascospores; seasons differ in the timing of ascospore release and phoma leaf spotting, and the subsequent timing and severity of stem canker development. 3. Timing of fungicide application; one or two fungicide sprays can give good control of the disease but inaccurately timed applications are ineffective. These results will be discussed in relation to strategies for management of phoma stem canker, including agronomic practices (e.g. sowing date), forecasting severity of epidemics and optimising fungicide use for effective disease control.

INTRODUCTION

Leptosphaeria maculans causes phoma stem canker of oilseed rape (canola, *Brassica napus*), resulting in losses up to £40M p.a. in the UK. *L. maculans* is a species complex with differences in biology between the aggressive A-group/Tox$^+$ and the less aggressive B-group/Tox^0. The pathogen survives the summer on crop debris, upon which pseudothecia develop. These release airborne ascospores, the primary inoculum, over several months from the autumn through to the spring (West *et al.*, 2001). Experiments in Australia showed that crops within 500 m of a large source of ascospores are particularly at risk (Barbetti *et al.*, 2000). At least 4 hours of wetness are required for spores to germinate and infect leaves, mainly by penetration of stomata (Huang *et al.*, 2001). Leaf lesions (phoma leaf spots) develop 7-14 days after spore release in typical autumn temperatures but this time can vary with the cultivar and age of the leaf (Poisson & Pérès, 1999). The fungus is able to grow through the vascular tissue of the leaves to enter the stem, where yield-damaging cankers develop (Hammond *et al.*, 1985). Infections of the earliest leaves are associated with the most severe crown cankers and the greatest yield losses (West *et al.*, 2001). Hence in the UK, proximity to inoculum, late sowing or early ascospore release, an aggressive pathogen population and inappropriate fungicide treatments are factors contributing to severe epidemics.

MATERIALS AND METHODS

Epidemiological differences between A and B groups

Plant colonisation by A and B-group *L. maculans* was investigated in winter oilseed rape field experiments at Rothamsted (harvested in 2000, cvs Lipton and Capitol; and 2001, cv. Apex). The ratio of large, pale leaf lesions, attributed as A-group, to smaller, dark lesions (B-group) was assessed in untreated plots each month, with identification confirmed by isolation from sub-samples. Stem samples were examined in the late spring/early summer. Cultures isolated from crown cankers and stem lesions were identified as A or B group by colony morphology and pigment production (West *et al.*, 2002a). Isolations were also made from pieces of tissue from the cortex, wood and pith of the crown region. After harvest, crop debris were collected from untreated plots, divided into stem base debris (including the taproot region and stems <50 mm above ground) and upper stem debris (>100 mm above ground), and incubated outdoors in separate trays. During the autumn and winter, small pieces of each debris type (size *c.* 10 x 5 mm) bearing pseudothecia were removed weekly and their ascospores were ejected onto water agar. Single ascospores were transferred to PDA and the resulting colonies identified. Leaf lesion data were also collected in a third season (2002) from cv. Apex.

Disease timing, yield loss and effect of fungicides

Ascospores were trapped with a Burkard spore sampler and identified by microscopy to give a continuous record of their daily numbers from August through to June. The spore sampler was surrounded by eight trays, each ≈ 0.5 m^2 and containing ≈100 infected stems, collected the previous season and incubated outdoors.

The epidemiology and control of phoma stem canker was investigated in the seasons harvested in 1998, 1999 and 2000 using winter oilseed rape cvs Capitol and Lipton at Rothamsted. Plots were sown in late August in four randomised blocks of five main plots, with each main plot split between the two cultivars. The sub-plots were 15 x 3 m, separated by 3 m guard-rows. Main plots received either no fungicides (control) or various fungicide regimes designed to produce different epidemics of phoma leaf spot (Table 1). The fungicide used was a mixture of difenoconazole (at 62.5 g a.i. ha^{-1}) plus carbendazim (at 125 g a.i. ha^{-1}) applied in water at 220 L ha^{-1}. The incidence and severity (number of leaves affected per plant) of leaf spotting was recorded each month from a sample of 25 plants per plot. Phoma stem canker (crown cankers and phoma stem lesions) was scored on a 0-5 scale (where 0 = uninfected, 1 <25% stem circumference girdled, 2 = 25-50% girdled, 3 >50% girdled, stem firm, 4 > 50% girdled, stem weak, 5 = plant dead or lodged). Plots were desiccated (diquat at 1.5 g L^{-1} a.i.) before harvest, combine harvested in mid-July and yields were taken. A further six experiments were done on cv. Apex at Rothamsted, ADAS Boxworth, Cambs and ADAS High Mowthorpe, N. Yorks. in harvest years 1999 and 2000, using the same fungicide treatment applied on four dates (4-6 week interval between sprays) from onset of phoma spotting in October/ November in a full factorial design (Gladders *et al.*, 2001).

The activity of the fungicide mixture used in the field experiment was investigated in controlled environment experiments. Leaves six and seven of 36 potted plants (cv. Lipton) were each inoculated with a drop of *L. maculans* ascospore suspension (2 x 10^3 spores mL^{-1}) at one point on each leaf (centrally, adjacent to the mid-rib). Plants were kept inside polyethylene chambers (100% relative humidity) for the first 3 days following inoculation.

Each week, subsets of six inoculated plants were temporarily removed and sprayed with the fungicide mixture at the concentration used in the field. Isolations on distilled water agar (DWA) were made from the inoculation site from three untreated plants and from three fungicide-treated plants (two leaves per plant) 1 day after the fungicide application and from another three treated plants 1 week after the fungicide application.

Table 1. Dates of fungicide applications in field experiments to study factors affecting phoma stem canker (*L. maculans*) on winter oilseed rape (cvs Capitol and Lipton) during three seasons at Rothamsted.

Fungicide regimes	Dates of applications each season		
	1997/98	1998/99	1999/2000
Control	none	none	none
A1	Oct, Nov, Feb	Oct	Oct
A2	Nov, Jan, Feb	Oct, Nov	Oct, Nov
A3	Oct, Nov, Jan, Feb	Oct, Nov, Jan	Nov, Mar
SF	Sept, Oct, Nov, Jan, Feb	Sept, Oct, Nov, Jan, Mar	Sept, Oct, Nov, Jan, Mar

RESULTS

Epidemiological differences between A and B groups

In 1999/2000 and 2000/2001, leaf spots were mainly caused by A-group *L. maculans* (66 to 88% A-group) but in 2001/2002 the B group was less rare and became more common than the A-group in late-winter and spring (52 to 58% A-group in October to January, 50 to 43% A-group in February to March). The A-group was predominant at the root and crown region (76% of samples), where it was found throughout the cortex, wood and pith. The B group was less common at the root and crown region and became rarer in samples from the wood and pith tissues (29% of samples were B-group in the cortex and wood, decreasing to 9% in the pith). From the upper stem region, the B-group was isolated at a similar frequency to the A-group (45% A, 55% B). Ascospores produced from debris of the root/crown were 94% A group, while from the upper stem40% were A-group and 60% B-group.

Disease timing, yield loss and effect of fungicides

Ascospores were detected in low numbers (<4 spores m^{-3} of air) in August and early September. Ascospore numbers increased in the autumn as pseudothecia on stubble from the previous harvest matured and the incidence of leaf-spotting increased *c.* 2 weeks after spore release. The fungicide mixture, delayed the onset of leaf spotting by 1 month in the autumn, and by 2 months in winter (there was a longer incubation period in cold weather, data not shown; West *et al.*, 2002b). The development of phoma leaf spotting in untreated plots differed between the seasons; in 1997/98 the incidence of affected plants reached 20% in late October with a maximum incidence of 31% in early December. In 1998/99, the incidence reached 20% in early/mid November with a maximum of 43% in mid-December. In 1999/2000 the incidence reached 20% in early October with a maximum of 84% in mid-October.

In controlled conditions, no colonies could be isolated from lesions on leaves treated with the fungicide 1 week after inoculation (Table 2). However, colonies were formed from all inoculation sites on leaves treated 3 weeks after inoculation, although these colonies had grown less on DWA after 8 days (13.7 mm) than those from untreated leaves (42.5 mm).

Table 2. Number of *L. maculans* cultures from oilseed rape leaves (cv. Lipton) inoculated with ascospores on 27 Sept 1999

Weeks post inoculation when fungicide applied	No. cultures isolated 1 day after fungicide application		No. cultures isolated 1 week after application
	Untreated leaves	Treated leaves	Treated leaves
1 (4 Oct)	6/6	0/6	0/6
2 (11 Oct)	6/6	3/6	2/6
3 (18 Oct)	6/6	6/6	3/6
4 (26 Oct)	6/6	5/6	(not tested)

The incidence of crown cankers in the spring was highest in untreated plots and increased throughout the spring. There was a trend for the incidence to be progressively lower in plots with an increasing number of fungicide applications (Figure 1).

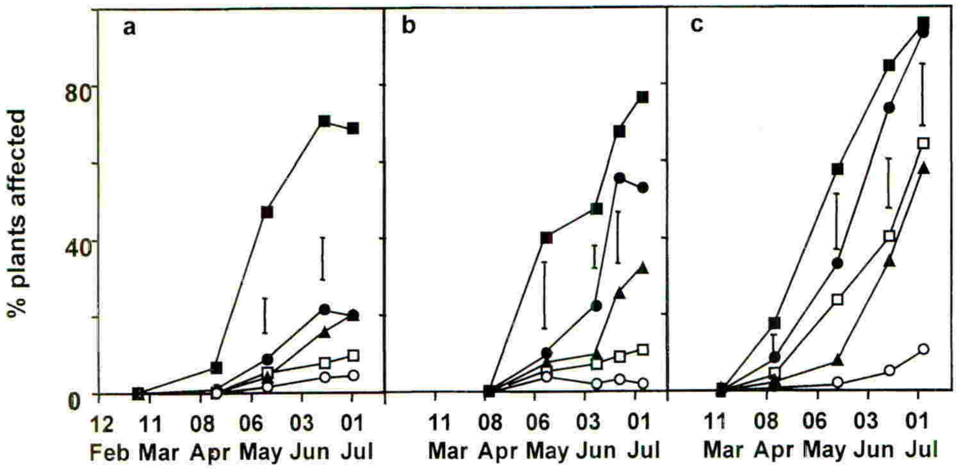

Figure 1. Crown canker incidence (% plants affected) with time in winter oilseed rape (mean of cvs Capitol and Lipton) grown under different fungicide regimes in 1998 (a), 1999 (b) and 2000 (c). Vertical bars represent LSDs where differences are significant ($P<0.05$). Untreated control ■, A1 ●, A2 ▲, A3 □, September to February (SF) O. Application dates for each fungicide regime are given in Table 1.

The severity of crown cankers on affected plants was also reduced by the use of fungicides (Figure 2). Statistically significant yield losses occurred when >50% of the stem circumference was girdled by crown canker by early July (severity score of 3 or more), which occurred only in untreated plots in 1997/98. Significant yield increases (0.69-1.03 t/ha) were recorded in all three factorial experiments in 2000 and at Rothamsted in 1999. Although crown canker severity in untreated plots differed between seasons, the rate of increase in severity with time was linear and did not differ between seasons, increasing by a score of 1 every 40 days. Hence, the relatively late leaf spotting epidemic of 1998/99 was associated with a late start to crown canker development, resulting in a lower final mean severity than in 1997/98 and no yield reduction. Stem lesions were more common in plots that did not receive fungicides after November (data not shown) indicating that they originated from infections of uppermost leaves from January onwards. These leaves originally appeared to belong to the rosette leaves but were raised to be above the true rosette leaves by internode extension from February onwards. Stem lesions did not become severe in any of the three seasons and were considered unlikely to have contributed to yield loss.

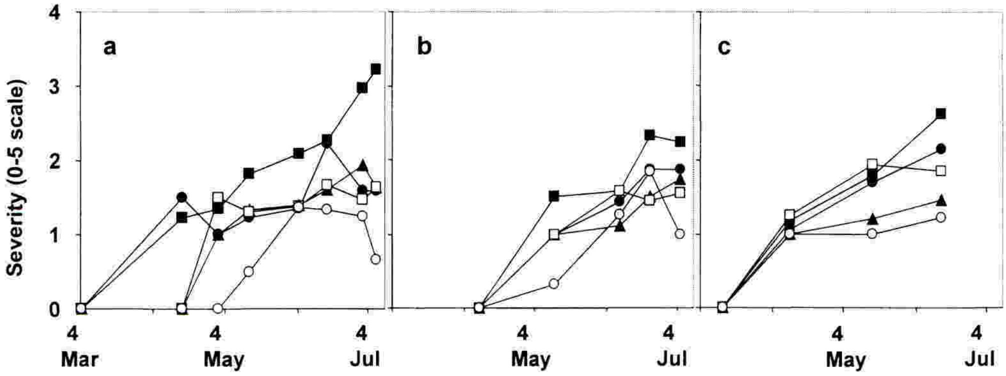

Figure 2. Severity of crown canker (0-5 scale) with time, in winter oilseed rape (mean of two cvs, Capitol and Lipton) grown under different fungicide regimes (A1, A2, A3, SF) in 1998 (a), 1999 (b) and 2000 (c). Symbols as for Figure 1.

DISCUSSION

Damaging crown cankers are caused predominantly by the A-group of *L. maculans*, while the rarer B-group tends to be confined to the cortex tissues and is more common on upper stem debris. Burial of the lignified tap root and crown material after harvest is therefore an important part of a control strategy as the main increase in leaf spotting was associated with spores released from stubble of the previous harvest. A low background concentration of ascospores, produced after rain in the summer, is thought to come from old stubble. The fungicide mixture used, did not usually eradicate leaf infections that were older than one week but reduced the fungal growth rate in treated leaves. This delay may be sufficient to prevent canker formation if the pathogen fails to reach the stem before leaf fall. On large plants, there is increased flexibility with spray timing and often improved disease control because long petioles delay or prevent stem infection. However, on small plants (less than 6 leaves) it would be essential to apply a fungicide before most leaf spotting appears to ensure that the fungus does not reach the stem. Early sowing to provide well-established plants (GS >1, 6) before the onset of phoma spotting is therefore a potentially valuable component of canker control. Further work on forecasting spore release and leaf spotting is continuing as part of the SAPPIO LINK–funded PASSWORD project, which aims to develop a decision support scheme for pests and diseases of winter oilseed rape. Furthermore, the indication here, that early leaf spotting leads to early canker development was shown by Sun *et al.* (2001) to be linked to thermal time (*c.* 1200 day degrees) following leaf infection. Fungicide regimes preventing all autumn and winter leaf infections were not the most economical control strategy. The aim is to prevent moderate or severe cankers that causes yield loss. In vigorous crops, the most economic control strategy may be a single foliar fungicide application, but two sprays are often required particularly if light leaf spot (*Pyrenopeziza brassicae*) control is also needed. Variation in the pathogen population is poorly understood and further work is continuing, aimed at improving strategies to exploit cultivar resistance in management of *L. maculans* in oilseed rape. In the short term, however, well-timed fungicide treatments and early crop establishment offer a cost-effective solution to damaging stem canker epidemics.

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Turnip rape (*Brassica rapa*) as a trap crop to protect oilseed rape (*Brassica napus*) from infestation by insect pests: potential and mechanisms of action

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ABSTRACT

The potential of turnip rape (*Brassica rapa*) for use as a trap crop to protect spring oilseed rape (*Brassica napus*) from infestations by pollen beetles (*Meligethes aeneus*) and seed weevils (*Ceutorhynchus assimilis*) was demonstrated in the field, and experiments investigating the mechanisms of its successful action initiated. Turnip rape plots developed more quickly, and became infested with pests earlier than those of oilseed rape. Pollen beetles remained on turnip rape until the oilseed rape was well past the damage-susceptible stage of yellow bud. Seed weevil numbers remained low on oilseed rape throughout the observation periods, being consistently higher on turnip rape. Results indicate that turnip rape has good potential as a trap crop to protect oilseed rape from these pests. Novel experiments conducted on a semi-field scale, and olfactometry studies suggest that the success of turnip rape as a trap crop is due to its earlier flowering with respect to oilseed rape and its more attractive odour. Computer simulations indicate that the trap cropping system could be effective on a whole-field scale. A model based on data from these experiments predicted less damage from pests in crops protected by a turnip rape trap crop than in oilseed rape monocultures.

INTRODUCTION

At Rothamsted Research, we are developing control strategies for insect pests of oilseed rape (*Brassica napus*) (OSR) that will reduce the current dependence on insecticides. Trap cropping strategies work on the principle that most pests show preferences for certain plant species, cultivars, or growth stages over others. These preferred plants can be used as 'trap crop' stands that attract the colonising pests away from the main crop, concentrating them on the trap plants where they can be killed or their populations maintained past the susceptible stages of the main crop.

Pollen beetles (*Meligethes aeneus*) and seed weevils (*Ceutorhynchus assimilis*) are two of the principal coleopteran pests of oilseed rape. Studies in mainland Europe have shown that turnip rape (*Brassica rapa*) (TR) is preferred by these pests and has potential for use as a trap crop to reduce damage to OSR crops (e.g. Hokkanen *et al.*, 1986). In this paper, we present the results of experiments designed to test the preferences of pollen beetles for TR and OSR in laboratory and semi-field scale experiments, attempting to define the mechanisms important in the success of this trap-cropping system. We also present the results of field trials, conducted over two years, to investigate the possibility of using spring-sown TR as a trap crop to protect spring OSR in UK conditions, using cultivars of OSR and TR available to UK farmers on the NIAB recommended lists.

MATERIALS AND METHODS

Linear track olfactometer

Dual-choice test experiments were performed using a linear track olfactometer bioassay (Cook *et al.*, in press) to determine if there were any preferences of pollen beetles for the odours of TR (cv. Agena) or OSR. Two cultivars of OSR were tested: Canyon, with a normal glucosinolate profile and Starlight, with lower proportions (Bartlet & Kiddle, unpublished data) of alkenyl glucosinolates (which catabolise to form the isothiocyanates to which crucifer-specialist insects are most attracted (Bartlet *et al.*, 1999)). Female pollen beetles (25) were introduced into the olfactometer, and the number turning left or right into one of two equal-flow airstreams (1 litre/min) that had been passed through test materials, was recorded for 15 minutes. Six experiments were conducted. In 1-3, the odour from flowering racemes (5 g) of Starlight, Canyon and TR, respectively, were tested against a blank air control (Whatman filter paper moistened with distilled water). In 4, the odour of Starlight was tested directly against that of Canyon, and in 5-6, the odour of Starlight and Canyon were tested against that of TR. For each experiment, 8 replicates were conducted using two olfactometers. Test odours were introduced into each chamber of the olfactometers (chamber 1 & 2), in each direction (left & right) to reduce the effects of any bias. Beetles were used only once, and floral material was replaced after every four replicates. The proportion of beetles turning left or right when a given odour was on the left or right was analysed in a 2×2 contingency table by a Pearsons χ^2 (GenStat, Release 4.2, VSN International Ltd., Oxford).

Polytunnel

The effect of plant growth stage on pest infestation of TR and OSR plants was investigated using a choice test conducted in a polytunnel arena. The polytunnel was 22.0 m long × 5.5 m wide × 2.5 m high and was clad in ultra violet inhibiting polythene. Six, 0.5 m diameter fans spanning the width of the tunnel were located behind a muslin screen at one end, and pulled air through the polytunnel at 0.5 m/s. Four OSR and four TR plants were placed in a row across the polytunnel with a 1 m gap between the two groups of plants. Pollen beetles (1,000) were released 5 m down-wind from the gap. The number of beetles present on each plant was recorded three hours after release. Four experiments were conducted. In experiment 1, both TR and OSR were flowering (growth stage (GS) 63-65 on the BBCH scoring scale of Lancashire *et al.* (1991)). In experiment 2, OSR was flowering and TR was in green bud (GS 53-57). In 3, OSR was in green bud and TR was flowering and in experiment 4, both were in green bud. For each experiment, 12 replicates were conducted, with three replicates performed in each direction (TR or OSR on the left or the right of the polytunnel) in morning and afternoon releases. Experiments were conducted in a randomised order each day. Pollen beetles were used once and plants were changed daily. Logistic regression was used to analyse the percentage of beetles attracted to the plants. A t-test on the logistic scale was used to test the difference between the percentage of beetles landing on TR plants and 50%. In analyses where the data exhibited overdispersion, a heterogeneity factor was estimated and used to model this variability in the responses of the beetles (see Cook *et al.*, in press).

Field plots

Turnip rape (cv. Agena) and OSR (cvs. Canyon and Starlight) were grown in twice-replicated plots (30 m × 30 m) in a randomised design. The plots were sown at the same time.

Infestation of the plots by adult pollen beetles and seed weevils was assessed twice in 1999, and three times when the experiment was repeated in 2000. On each occasion, the growth stage of the plots was recorded, and the number of beetles and weevils on the main raceme of 50 plants selected at random from each plot was recorded by beating each raceme onto a tray. The mean numbers of adult beetles and weevils per plant were calculated for the three plant types and compared.

Modelling the expected outcome from a turnip rape protected oilseed rape crop

To predict the effect of adding a TR trap crop to an OSR cropping system, the population dynamics of a hypothetical pollen beetle population were simulated using a spatially explicit individual-based simulation model (see Potting *et al.*, 2002), incorporating the results of the behavioural experiments in this study. Two types of trap crop border were simulated: (1) TR in flower, detected by the beetles principally by vision and (2) TR in bud, detected by the beetles principally by olfactory cues (i.e. upwind oriented orientation). For each simulation scenario, the mean expected damage in the crop was calculated from 40 simulation runs.

RESULTS & DISCUSSION

Linear track olfactometer

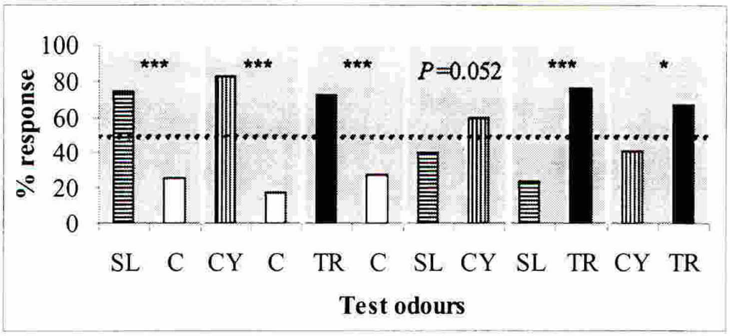

Figure 1. Responses of pollen beetles to oilseed rape and turnip rape odours in six dual-choice test experiments in a linear track olfactometer (SL = Starlight oilseed rape; CY = Canyon oilseed rape; TR = turnip rape; C = control; * $P < 0.05$; *** $P < 0.001$)

Pollen beetles were attracted to the odour of both cultivars of OSR and TR when these were presented against a blank air control (Figure 1). Previous studies have shown that pollen beetles are attracted to the odour of OSR in an olfactometer (detailed in Cook *et al.*, in press), but this is the first to demonstrate their attraction to TR odour. The odour of TR was preferred over the odours from both OSR cultivars by the beetles (Figure 1). Results from these laboratory studies indicate that TR has potential as a trap crop to protect OSR from pollen beetles as it has a more attractive odour.

More beetles responded to the odour of Canyon than Starlight in the choice test ($P = 0.052$) (Figure 1). The use as a main crop of OSR cultivars such as Starlight, that due to their glucosinolate content may be less attractive than other cultivars to crucifer-specialist insects (Bartlet *et al.*, 1999), may enhance the success of TR trap cropping systems by accentuating the difference in preference between the TR trap crop and the main crop.

Polytunnel

When both plant species were flowering, there was no difference in the proportion of pollen beetles that infested OSR or TR plants (Table 1). When one species was in flower and the other was in green bud, significantly more beetles were present on the flowering plants, irrespective of species (Table 1). When both OSR and TR were in green bud, TR was significantly more infested than OSR (Table 1). In each of the four experiments there was no evidence of experimental bias due to plant position or time of day.

These semi-field experiments predict that in a pre-flowering trap cropping system, beetles would infest TR more than OSR, and if TR develops quicker than OSR, would continue to do so until the OSR itself begins to flower. These results support the view that visual cues are important in host-location by the pollen beetle (e.g. Free & Williams, 1978) and highlight the requirement for precise growth/flowering phenology of the two species relative to each other for the success of a TR trap cropping system for OSR.

Table 1. Percentage pollen beetles present on oilseed rape (OSR) and turnip rape (TR) plants at different growth stages (flower or bud)

Experimental treatments	% beetles present		n	*P*
	OSR	TR		
OSR Flowers vs. TR Flowers (*)	48	52	1256	0.51
OSR Flowers vs. TR Buds (*)	92	8	984	<0.001
OSR Buds vs. TR Flowers	6	94	304	<0.001
OSR Buds vs. TR Buds	29	71	189	<0.001

(*) Analysis adjusted for overdispersion

Field plots

In both years, the plots of TR developed more quickly (Table 2) and became infested with pollen beetles (Figure 2 a&c) and seed weevils (Figure 2 b&d) earlier than those of OSR. There was little difference between the numbers of pollen beetles or seed weevils infesting Starlight and Canyon varieties of OSR (Figure 2), indicating that the lower proportions of alkenyl glucosinolates in Starlight had little effect on pest populations in the field.

Populations of pollen beetles increased as the season progressed, with similar profiles for both years (Figure 2 a&c). The number of beetles on TR remained relatively constant, but increased on OSR as it began to flower (Figure 2 & refer to Table 2). The number of beetles was significantly greater on TR than OSR until the final assessment in both years (Figure 2 a&c). At this time, the TR had only a few secondary racemes in flower while the OSR main racemes were flowering (Table 2). Therefore, the beetles were maintained on the TR plots past the damage-susceptible stage of yellow bud of the OSR plots. These results support the predictions made from the semi-field experiments and demonstrate on a field-scale that TR has good potential for protecting OSR against pollen beetles by reducing their populations to below spray threshold levels (Lane & Walters, 1993) during the susceptible stages.

Table 2. Growth stage of oilseed rape and turnip rape plots during pest assessments made in 1999 and 2000

Year	Assessment	Date	Growth stage (BBCH code, Lancashire *et al* (1991))	
			Oilseed rape	Turnip rape
1999	1	June 8th	51-57 (green bud)	61-63 (early flowering)
	2	June 15th	60-61 (early flowering)	71 (1° raceme pods; 2° racemes mid-flower)
2000	1	June 14th	50 (early green-bud)	51-57 (green bud)
	2	June 21st	59-60 (early flower)	65 (full flower)
	3	10th July	65 (full flower)	79 + (mostly pods)

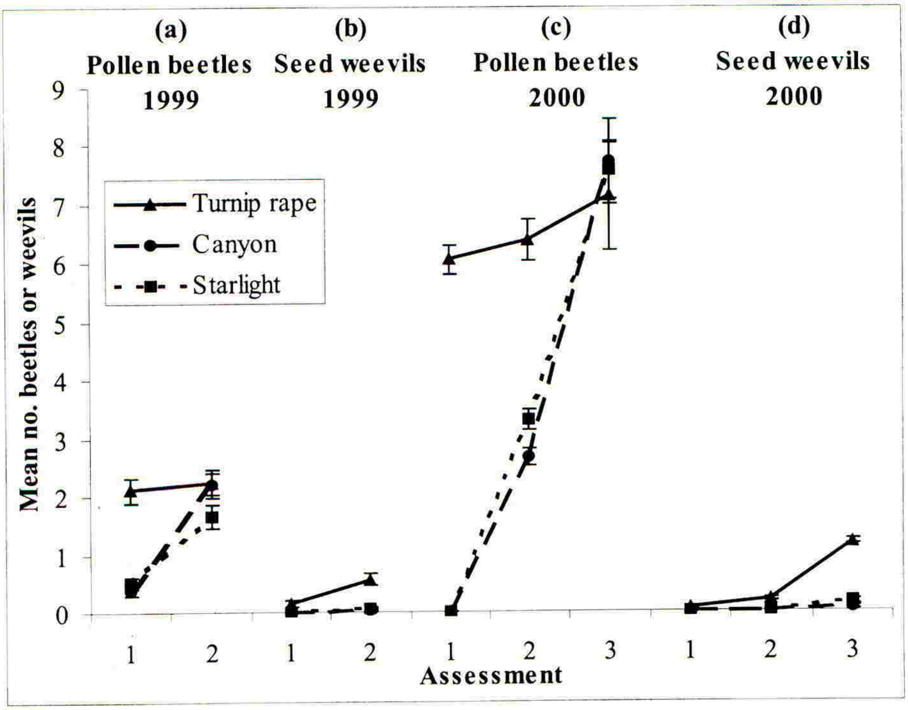

Figure 2. Mean (± SE) number of pollen beetles and seed weevils per plant on turnip rape (cv Agena) and oilseed rape cultivars (Canyon & Starlight) in 1999 and 2000

Populations of seed weevils were generally greater in TR than OSR plots (Figure 2 b&d). Their numbers increased on TR plots as the season progressed but remained low on OSR. However, numbers never reached spray-threshold levels of 2 weevils per plant (Lane & Walters, 1993). Unlike pollen beetles, seed weevils remain in the crop past flowering, as they lay their eggs in the pods. Assessments were discontinued before the OSR ceased flowering and populations may have increased on these plots after assessments ceased. Future experiments with assessments extended to the podding stages of OSR are required to fully support the potential of TR as an effective trap crop to protect OSR from seed weevil damage.

Modelling the expected outcome from a turnip rape protected oilseed rape crop

In computer simulations, the addition of a TR trap crop border reduces the herbivore damage in the OSR crop compared to a monoculture (ANOVA, P<0.001; Figure 3). The TR border is more effective when it is flowering than when it is in bud with respect to the main crop (Figure 3). These simulated field-scale results indicate the potential effectiveness of this approach in modern cropping systems. Future experiments conducted on a whole-field scale will test whether a TR trap crop border can reduce pest damage in protected OSR crops in practice, and explore any additional benefits the trap crop may bring through effects on the natural enemies of OSR pests.

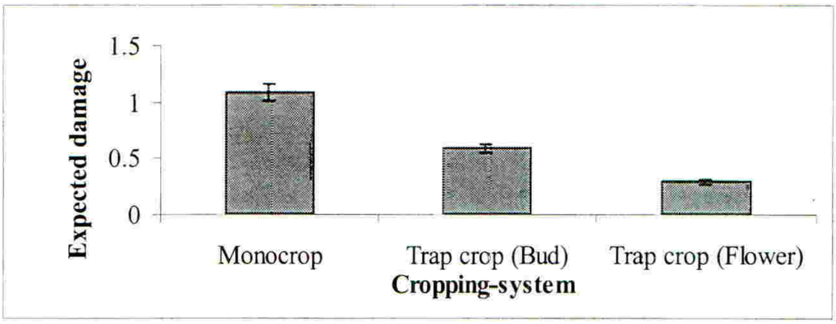

Figure 3. Mean (± SD) expected damage (herbivore days in the crop (Potting *et al.*, 2002)) to the main crop in different cropping systems after 50 days

ACKNOWLEDGEMENTS

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Efficacy of single and two-way fungicide seed treatments for the control of metalaxyl-resistant strains of *Plasmopara halstedii* (sunflower downy mildew)

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ABSTRACT

Control of the metalaxyl resistant strain of *Plasmopara halstedii* (causal agent of sunflower downy mildew) was studied in greenhouse and field experiments over a three-year period. Of 30 fungicides investigated, only azoxystrobin and fenamidone showed good efficacy as seed treatments for control of systemic symptoms initiated by root infection. Four fungicides displaying lesser disease control (ethaboxam, fluazinam, mancozeb, and zoximide) were tested as co-fungicides with azoxystrobin and fenamidone. None of the fungicides, either used alone or in combination, gave complete control under natural or inoculated field conditions, in contrast to the total control previously experienced with metalaxyl. Disease incidence on all fungicide treatments increased from 2 wk to 7 wk after emergence, suggesting that the fungicide/rates tested were more fungistatic than fungicidal. Fenamidone was the most effective fungicide used singly, and fenamidone combined with either ethaboxam or zoximide was the most effective combination. Greenhouse tests of fungicide seed treatments were not effective at predicting field response of the best treatments, despite several modifications. The greenhouse tests, however, were satisfactory to identify ineffective fungicides. None of the three fungicides likely to be registered as sunflower seed treatments in the United States, namely azoxystrobin, fenamidone, and zoximide, displayed any potential phytotoxicity or seed safety concerns in preliminary laboratory and field emergence tests, intended to run for two years.

INTRODUCTION

Downy mildew of sunflower *(Plasmopara halstedii*) has been controlled for the last 15 years largely through the use of fungicide seed treatments, primarily metalaxyl and the active stereoisomer, mefonoxim. In 1995, tests of 19 *P. halstedii* isolates from the US, Hungary, China and Argentina revealed no isolates which were able to infect seeds treated with 5 g a.i. metalaxyl/ 100 kg seeds (Gulya & Viranyi, 1995). Resistance to metalaxyl in *P. halstedii* was first noted in France in 1995 and in the United States in 1998 (Gulya *et al.*, 1999). In the north central region of the United States, 91% of nearly 200 isolates of *P. halstedii* collected in 1998 and 1999 were tolerant of metalaxyl. Although the apparent resistance of bulk field isolates appeared variable, once spores were collected from metalaxyl-treated seedlings, all subsequent inoculations produced 100% infection on metalaxyl treated seeds. The metalaxyl tolerance is rate-insensitive, with no control observed on rates ranging from 63 to 960 g a.i. metalaxyl/100 kg seeds. In the U.S., sunflower hybrids with genetic resistance to downy mildew were not available when metalaxyl-resistance was first observed, so it became imperative to find new fungicide seed treatments to control the metalaxyl-resistant strains. This study commenced with greenhouse evaluations of candidate fungicides (1999-2000), progressed to field testing under natural infection (2000), and culminated in inoculated field trials (2001-2).

In preliminary greenhouse tests, nearly 30 fungicides were evaluated as seed treatments, each at rates from 31 to 2500 g ai/100 kg (Gulya *et al.*, 1999). The fungicides tested included older, non-systemic fungicides (captan, chloroneb, chlorothalonil, copper hydroxide, copper tallate, etridiazole, mancozeb, thiram, and streptomycin), newer, systemic fungicides, most of which have activity against Oomyctes (azoxystrobin, cymoxanil, dimethomorph, fludioxinol, fosetyl-Al, kresoxim methyl, hymexanol and propamocarb) and newer, experimental fungicides with reported activity versus Oomycetes (Actigard, Bayer IFK 916, DuPont KP-481 and KZ-165, ethaboxam, famoxadone, fenamidone, fluazinam,, Tomen Agro SZX-722, and zoximide). Of these compounds, the fungicides with the best efficacy were azoxystrobin and fenamidone used individually, with fluazinam, mancozeb and zoximide the best co-fungicides.

MATERIALS AND METHODS

Seed treatment and greenhouse evaluation method

Sunflower seeds (oilseed hybrid 'Cargill 270') were treated either by hand mixing in 100 g quantities in Erlenmeyer flasks for greenhouse tests, or in 500-1000 g quantities in a Hegge treater for field experiments. In both cases, the volume of aqueous fungicide was 2% of the seed volume. For greenhouse evaluations, treated seeds were planted in 25 x 25 x 4 cm plastic flats filled with a sand/perlite (2:1, v/v) mixture, and grown in a greenhouse maintained at 22-25°C with a 16 h photoperiod. On the 3rd day after planting, the flats were drenched with 100 ml of a suspension of *P. halstedii* zoosporangia (2×10^4/ml). The inoculation was repeated for three additional days. Two weeks after planting (11 days after the first inoculation), the flats were transferred to a room maintained at 16°C, misted, and held overnight to induce sporulation. Diseased plants had profuse sporulation on cotyledons and true leaves, and were removed from the flats. The remaining, apparently uninfected plants were grown an additional two wk in the greenhouse. The plants were carefully dug up, the roots rinsed in tap water, cut off at ground level, and placed in plastic boxes lined with wet blotter paper. The boxes were held in the dark at 16°C for 48 h and the root examined under a dissecting microscope at 25-50X for the presence of zoosporangia on the root surfaces. Total downy mildew infection was calculated as the percentage of plants showing foliar symptoms and the percentage of plants showing root colonization. The test was further modified by (1) planting individual seeds in nursery "cell packs" and (2) by using a sandy loam soil rather than the sand/perlite mixture.

Field evaluation methods

Field experiments in 2000 were planted at 24 locations in five states and in Manitoba, Canada at sites presumed to have soilborne *P. halstedii*. Four replications of single row plots were seeded as early as possible to expose the seed to cool, wet soil conditions conducive to root infection (Gulya *et al.*, 1997). A total of 24 treatments were tested, of which 12 are listed in Table 1. Field experiments in 2001 were conducted at five locations in Minnesota and North Dakota, all of which were set up with drip irrigation systems. The drip hose had emitters spaced every 20 cm and supplied 45 l water/min/100 m. Immediately after planting all rows were irrigated to facilitate rapid and uniform emergence. Seeds were dug up daily to examine germination, and the rows were inoculated when radicle length reached 1 cm (4 - 9 days after planting). The rows were pre-irrigated for two hours, the inoculum applied, and then an additional 4 hours of water were applied. Inoculum, freshly collected from greenhouse grown seedlings, was applied at 10 x 10^9 zoosporangia/ m^2, assuming an irrigated width of 15 cm. Disease evaluations were made 2 wk after inoculation and continued at biweekly intervals until 7 wk after inoculation. A total of 12 field experiments were conducted in 2001, with from five to 24 treatments evaluated per experiment. The results of one experiment repeated at five locations are listed in Table 2.

Field experiments in 2002 were evaluated for both disease control and any potential phytotoxicity. Twenty-six oilseed and confection sunflower hybrids were treated with the three fungicides most likely to be registered in the United States, and at 2X rate. The control (and background treatment on the six fungicide treatments) consisted of mefonoxim (16 g a.i.), fludioxinol (5.3 g a.i./100 kg seed), which was intended to protect seedlings against soilborne diseases other than downy mildew, and the insecticide pirimophos-methyl (0.85 g a.i./100 kg seed for protection against stored grain insects. Seeds were treated in January, 2002 for a seed safety study intended to last 24 months. Seeds were stored at room temperature and a 38°C in an attempt to hasten any potential phytotoxicity. At three month intervals, 400 seeds of each hybrid/fungicide treatment (182 total) were tested in standard warm germination tests, and in an accelerated aging tests where the seeds were held at 42°C for 72 h at 100% RH prior to the germination test. Field emergence was tested at four sites in 2002 and will be tested in 2003, after 16 months in storage. Emergence counts and phytotoxicity notes were made at 2, 3 and 4 wk after planting. Disease control was assessed at a single inoculated field site, using the procedures described previously.

RESULTS

Preliminary greenhouse tests (data not shown) revealed that azoxystrobin and fenamidone had the highest efficacy against root initiated systemic infection by *P. halstedii*. Ethaboxam, fluazinam, mancozeb and zoximide, with lesser efficacy and different modes of action, were considered the best candidates as co-fungicides with either azoxystrobin or zoximide. Of 24 field locations in fives states planted in 2000, only two plots had enough natural infection to evaluate disease control (primarily due to lack of waterlogged soils after planting). Disease evaluations changed drastically, from the initial observations 3 wk after planting until 7 wk, on nearly all fungicide treatments, while disease incidence remained stable on the untreated and metalaxyl treatments. As an example, the best treatment, fenamidone @ 125 g a.i., had 1 and 0 % infected plants at 3 wk in fields 1 and 2, respectively, which increased to 16 and 3% by 7 wk. Plants at 7 wk after emergence had 5 to 7 pair of leaves, with the lower leaves healthy, intermediate leaves displaying minimal chlorosis bordering midveins, and uppermost leaves displaying increasingly more area with chlorosis. This delayed onset of symptoms on fungicide treated seedlings suggested the chemical/rates being tested were fungistatic rather than fungicidal. There was also a major discrepancy between greenhouse results and field results (Table 1). Even the best treatment (fenamidone @ 200 g a.i.), which gave 100% disease control in greenhouse tests, had from 3 to 16% infected plants in the two field trials. All fungicides, either used singly or in combinations, displayed a similar lack of correlation between greenhouse and field results. This discrepancy prompted us to revise the greenhouse evaluation method by growing apparently healthy seedlings an additional 2 wk, incubating washed roots at 16°C for 48h in moist chambers, and then examining the roots for sporulation under a dissecting microscope at 25X.

Table 1. Efficacy of fungicide seed treatments for control of sunflower downy mildew in greenhouse trials and field trials under natural infection in 2000.

	Downy Mildew Incidence (%)		
Fungicide/Rate (a.i./100 kg seed)	Field 1	Field 2	Greenhouse
Untreated	83	41	70
Metalaxyl @63 g	78	42	71
Azoxystrobin @ 63 g	45	13	0.4
Azoxystrobin @ 125 g	46	11	1.2
Fenamidone @ 63 g	29	4	1
Fenamidone @ 125 g	16	3	0
Zoximide @ 250 g	48	14	8
Zoximide @ 375 g	38	11	6
Zoximide @ 500 g	40	6	5
Azoxystrobin @ 38 g + zoximide @ 63 g	49	10	1.7
Azoxystrobin @ 38 g + zoximide @ 125 g	35	7	0.6
LSD (P=0.05)	12	8	4

In 2001, a total of 12 field trials were planted, all using drip irrigation to apply an artificial inoculum of zoosporangia. All trials developed disease except one in which the diseased leaves used for inoculum had been stored at 4°C for 2 wk. The remaining trials, which were planted from May 10 to Aug. 15, had from 18 to 90% infection on untreated seeds, with the lower levels of infection occurring on plantings made in the middle of the summer. Disease incidence in five early planted trials is shown in Table 2. The best treatment, averaged across the five sites, was the combination of fenamidone + ethaboxam (with 7% infection), followed closely by fenamidone (125 g), and the combination of azoxystrobin and ethaboxam, neither of which were significantly different. Since ethaboxam was not likely to be registered soon in the United States, emphasis in future tests was placed upon the three products likely to be registered, namely azoxystrobin, fenamidone, and zoximide. Results from greenhouse trials again failed to accurately predict field response of the fungicides, despite the use of root examinations of 4 wk old seedlings. A further modification of the greenhouse test was added during the winter of 2001, which entailed planted single seeds in nursery "cell packs" rather than planting 40 seeds in an open flat, and also by using a sandy loam soil rather than the sand/perlite mixture.

In another series of inoculated field experiments in late summer, 2001 (Table 3), the variation in field response was evident. Under moderate disease incidence in field 1, the combination of azoxystrobin and zoximide had the lowest disease incidence (3%), while this same treatment in field No.2 had the highest disease (31%). Averaged over both sites, the lowest disease incidence was observed on both rates of the fenamidone + ethaboxam combination, and the fenamidone + mancozeb combination. In comparison, a genetically resistant hybrid used as a control exhibited no disease in either field.

In 2002, one inoculated field trial was planted in which 26 hybrids were evaluated with 3 fungicide combinations at 1X and 2X rates (Table 4). Disease incidence again changed drastically from the initial to the final rating. At 2 wk after inoculation, there was no significant difference in disease between the respective 1X and 2X rates of the three treatments, while at 4 wk, the differences were significant. The best fungicide in this single field trial was fenamidone at 250 g a.i. No differences in laboratory germination in warm germination tests or accelerated aging tests, nor in field emergence, were noted between any

Table 2. Efficacy of fungicide seed treatments for control of sunflower downy mildew inoculated field and greenhouse trials in 2001.

Fungicide/Rate (a.i/100 kg seed)	Downy Mildew Incidence (%)						
	Greenhouse Test	Field Trial No. 1	2	3	4	5	Av.
Untreated	99	24	22	63	92	96	64
Metalaxyl @ 63 g	–	20	14	66	90	90	56
Azoxystrobin @ 125 g	8	20	10	19	45	45	29
Azoxystrobin @ 188 g	10	6	8	13	31	26	19
Fenamidone @ 125 g	13	3	3	8	37	18	14
Fenamidone @ 188 g	5	5	3	6	25	17	12
Azoxystrobin @ 63 g + zoximide @ 125 g	1	4	7	17	48	33	24
Azoxystrobin @ 125 g + zoximide @ 250 g	1	8	6	18	30	31	20
Azoxystrobin @ 63 g + ethaboxam @ 250 g	3	3	5	7	30	15	13
Fenamidone @ 63g + zoximide @ 125 g	6	1	3	15	40	24	19
Fenamidone @ 125 g + zoximide @ 250 g	3	3	5	14	33	28	19
Fenamidone @ 63 g + ethaboxam @ 250 g	1	3	2	5	12	12	7
LSD (P=0.05)	3	10	6	11	12	10	5

Table 3. Efficacy of fungicide seed treatments compared to genetic resistance for control of sunflower downy mildew in inoculated field trials in 2002.

Fungicide/Rate (a.i./100 kg seed)	Downy Mildew Incidence (%)		
	Field 1	Field 2	Average
Untreated	24	53	39
Azoxystrobin @ 125 g + zoximide @ 250 g	3	31	17
Fenamidone @ 125 g + zoximide @ 250 g	7	17	12
Fenamidone @ 63 g + ethaboxam @ 250 g	6	9	8
Fenamidone @ 125 g + ethaboxam @ 250 g	9	7	8
Fenamidone @ 125 g + mancozeb @ 250 g	12	10	11
Genetic resistant hybrid	0	0	0
LSD (P=0.05)	3	4	3

DISCUSSION

Metalaxyl resistant *P. halstedii* is widespread in the sunflower growing areas of north-central United States, and genetically resistant hybrids are only slowly becoming available.

Table 4. Efficacy of fungicide treated sunflower seed for the control of downy mildew in one inoculated field trial in 2002, averaged over 26 hybrids, and emergence in four, non-diseased fields.

Fungicide/Rate (a.i./100 kg seed)	Disease Incidence 2 wk	Disease Incidence 4 wk	Range over 26 hybrids	Emergence (%)
Metalaxyl @ 63 g	24	51	36 to 77	72
Azoxystrobin @ 63 g + zoximide @ 125 g	10	38	17 to 62	72
Fenamidone @ 63 g + zoximide @ 125	6	21	11 to 45	70
Fenamidone @ 125 g	3	15	4 to 40	71
Azoxystrobin @ 125 g + zoximide @ 250 g	8	35	13 to 66	71
Fenamidone @ 125 g + zoximide @ 250 g	4	16	2 to 29	70
Fenamidone @ 250 g	2	8	2 to 18	72
LSD (P=0.05)	3	4		NS

Evaluation of isolates from European countries (Hungary, Bulgaria, Spain), and South Africa has shown that metalaxyl resistance occurs in other countries, either through natural selection of the native fungal population, or by introduction from other countries. It is likely that genetic resistance will not be incorporated into commercial hybrids in all countries, making an effective fungicide seed treatment a necessary alternative control option. Three fungicides (azoxystrobin, fenamidone, and zoximide) are, or will be registered in the U.S. for use against potato late blight, and thus will be likely to be registered for use as sunflower seed treatments thereafter. After multiple, inoculated field trials in two years, it appears that fenamidone, used alone at rates of 125 g a.i. or higher/100 kg seed, offers the best control against *P. halstedii*. In an attempt to curtail the development of fungicide resistance, and to minimize costs, it is wise to continue evaluating fungicide combinations and to urge both the seed and chemical industry to pursue this avenue.

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