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Susceptibility of winter rape cultivars to fungal diseases and their response to fungicide application

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Abstract: The productivity of winter oilseed rape cultivars is limited by various pests and diseases. The aim of this study was to determine the differences in the susceptibility of Lithuanian-grown winter rape cultivars to main fungal diseases, their yield performance, and their response to the fungicide application. A 2-factorial field experiment was carried out at the Institute of Agriculture of the Lithuanian Research Centre for Agriculture and Forestry over 3 growing seasons (2007/08, 2008/09, and 2009/10). The influence of fungicide application and cultivar (15 cultivars during the first and second seasons, and 6 cultivars during the third season) on the incidence and severity of phoma stem canker, alternaria black spot, and sclerotinia stem rot, as well as on the seed yield and thousand-seed weight, was evaluated. Experimental findings indicated that the incidence and severity of fungal diseases varied considerably among the growing seasons. The incidence of basal phoma stem canker was significantly influenced by the cultivar during all 3 growing seasons; however, significant effect of the cultivar on the mean severity of basal canker was obtained only in 2009. Fungicide spray applications significantly decreased the incidence and severity of fungal diseases and significantly increased the seed yield of winter rape. Significant and positive yield response to the fungicide application was obtained for almost all tested cultivars during the first 2 seasons. Cultivars that were higher yielding without treatment (Insider, Sunday, Monalisa, Rally, and Titan) showed lower response to the fungicide application. The use of fungicides demonstrated the highest economic return from the cultivars Lirajet, Casino, Siska, Liprima, and Valesca.

Key words: Diseases, economic benefit, fungicides, seed yield, winter rape

1. Introduction

The productivity of oilseed rape (*Brassica napus* L. var. *oleifera*) is limited by various fungal diseases (Sharma and Kolte, 1994; Bardin and Huang, 2001; Pearse et al., 2001; West et al., 2001; Fitt et al., 2006a). Phoma stem canker is a very common and damaging disease of oilseed rape in many countries (Huang et al., 2005; Fitt et al., 2006a, 2006b). The complex of 2 fungus species, *Leptosphaeria maculans* and *L. biglobosa*, causes phoma stem base or crown canker and upper stem lesions (West et al., 2001). Differences in disease severity occur between sites and seasons and may be partially attributed to the differences in weather conditions favourable for infection of leaves by ascospores of the fungus (West et al., 1999, 2001). Different seasonal types (winter, spring) and cultivars of oilseed rape also differ in the susceptibility to this disease (Delourme et al., 2006). Seed losses from phoma stem canker usually reach 10%, but in some years they can reach 30%–50% (Hall et al., 1993; Zhou et al., 1999). Alternaria black spot

(*Alternaria brassicae*) and sclerotinia stem rot (*Sclerotinia sclerotiorum*) are also very important fungal diseases of oilseed rape (Hong et al., 1996; Koch et al., 2007).

Cropping practices affecting the severity of phoma stem canker include crop rotation, stubble management and tillage, sowing date, plant density, and fertiliser use (Aubertot et al., 2006). Effective control of phoma stem canker can also be achieved by foliar fungicide sprays in the autumn; however, optimal timing of such applications is very important (West et al., 2002; Aubertot et al., 2006; Gladders et al., 2006). Useful effects may also be achieved through the choice of resistant cultivars (Delourme et al., 2006; Fitt et al., 2006a). The control of alternaria black spot and sclerotinia stem rot, however, is based on the application of fungicides (Kutcher and Wolf, 2006; Stammler et al., 2007; Brazauskienė et al., 2011a). Oilseed rape breeding is not conducted in Lithuania and all cultivars of this crop grown by farmers have been developed abroad. As a result, their susceptibility to the fungal diseases caused by

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the local populations of the fungus may differ. However, there is little information about the response of the current Lithuanian-grown oilseed rape cultivars to fungal diseases. The information from the Variety Testing Centre about the disease resistance of the cultivars included in the list of cultivars recommended for cultivation in Lithuania is very limited. In Lithuania, winter hardiness of winter rape cultivars is the most important indicator; therefore fungicide sprays (mainly triazoles, possessing plant growth regulation properties (Pits et al., 2008)) are primarily applied in autumn (at growth stage (GS) 15–16) for winter hardiness improvement rather than disease control. Sclerotinia stem rot has become a major problem during recent years and the farmers routinely use fungicides at the flowering stage. However, information on the efficacy of fungicide sprays against complexes of fungal diseases is not available. As a result, the aim of this study was to determine the differences in the susceptibility of winter rape cultivars to fungal diseases, their yield performance, and their response to fungicide application (at GS 15–16 and GS 63) in field trials.

2. Materials and methods

2.1. Field trial conditions

Field experiments were carried out in the experimental field of the Institute of Agriculture, Lithuanian Research Centre for Agriculture and Forestry. The experiments were set up with 15 cultivars of winter oilseed rape (*B. napus*) during the 2007/08 and 2008/09 growing seasons, and with 6 cultivars during the 2009/10 growing season.

Conventional soil tillage and drilling techniques were applied. For the 2007/08 season, winter rape was sown after the fallow. Before sowing, the soil was fertilised with potassium and phosphorus at the dose rates of 100 and 60 kg ha⁻¹, respectively. During presowing the field was also sprayed with the herbicide Triflurex 48 EC (a.i. trifluralin, 480 g L⁻¹) at 2.5 L ha⁻¹. Shortly after application, the herbicide was incorporated into the soil with a Kongskilde germinator, and the field was sown with winter rape on 21 August. In the spring, on 1 April, winter rape was fertilised with ammonium sulphate at a rate of 140 kg ha⁻¹ of nitrogen. To control pollen beetle (*Meligethes aeneus*), insecticide Karate Zeon 5 CS (a.i. λ-cyhalothrin, 50 g L⁻¹) was used (0.15 L ha⁻¹) at GS 57.

For the 2008/09 and 2009/10 seasons the field for winter rape was prepared after winter wheat harvesting. The field was shallowly (5–8 cm) harrowed to promote germination of weed and volunteer wheat seeds. Approximately 2 weeks later the field was ploughed with a reversible plough in combination with a soil compactor. Prior to sowing, the soil was fertilised with nitrogen, phosphorus, and potassium at the dose rates of 20, 60, and 120 kg ha⁻¹, respectively.

Winter rape was sown at the end of the second 10-day period of August. A day later, herbicide Brasan 540 EC (a.i. dimetachlor + clomazone, 500 + 40 g L⁻¹) was sprayed at a dose rate of 2.0 L ha⁻¹. In the spring of 2009 and 2010, nitrogen (ammonium nitrate) was applied twice at 90 and 60 kg ha⁻¹, respectively. To prevent sulphur deficiency and to control pollen beetle, the field was sprayed (GS 55) with dissolved ammonium sulphate at a dose rate of 20 kg ha⁻¹ in a mixture with insecticide Decis Extra 100 EC (a.i. deltamethrin, 100 g L⁻¹) at 0.06 L ha⁻¹.

In all experimental years the seeds were treated with Cruiser OSR 322 FS at 11.25 kg t⁻¹ (a.i. tiametoxam + fludioxonil + metalaxyl-M, 280 + 8 + 33.3 g L⁻¹) and sown at a rate of 80 viable seeds per square metre at a sowing depth of 2–3 cm. Sowing rate was determined for each cultivar by using thousand-seed weight (TSW) and seed germination data. The plots of each cultivar were sown with a trial drilling machine, Hege-80 (Wintersteiger, Austria).

2.2. Experimental design

The experimental design was a split plot in 4 blocks (replications) with 2 main plots (untreated vs. fungicide-treated), containing 15 cultivar subplots (2007/08 and 2008/09 cropping seasons) or 6 cultivar subplots (2009/10 season). The following commercially grown cultivars of winter rape were selected for this study: Celsius, Casino, Insider, Libea, Liprima, Lirajet, Silvia, Siska, Sunday, Valesca, Kasimir, Kronos, Monalisa, Rally, and Titan. Amongst the cultivars, Silvia, Kronos, and Sunday are characterised by resistance to phoma stem canker. Cultivar Celsius was used as a control. The seeds for the trials were obtained from a seed production company (Dotnuvos projektai, Lithuania). The plot size of each cultivar was 30 m² (3 × 10 m). In the untreated main plot, fungicides were not used, and in the fungicide-treated main plot, winter rape cultivars were sprayed twice with fungicides to control the main diseases. The first application was performed in autumn at GS 15–16 with fungicide Juventus 90 (a.i. metconazole, 90 g L⁻¹) at 0.75 L ha⁻¹ (to control basal phoma canker), and the second fungicide application was performed at GS 63 using Cantus 50 WG (a.i. boscalid, 500 g kg⁻¹) at 0.5 kg ha⁻¹ (to control sclerotinia stem rot, phoma upper stem lesions, and alternaria black spot). Fungicide applications were performed with a compressed air-powered Hardi-type sprayer using Hardi flat-fan nozzles (types 4110–20). The spraying pressure was 4.0 bars per nozzle. The fungicide-treated plots received 400 L ha⁻¹ of spray solution each time.

2.3. Disease and yield evaluation

Observations of phoma stem canker (basal canker and upper stem lesions) were made before harvesting (GS 85). Plant samples (30 plants per plot) were collected (15 consecutive plants in 2 rows) and analysed. The

mean disease incidence (% of plants with basal canker and % of plants with upper stem lesions) was calculated. The severity of basal canker was assessed by sectioning the plants at the crown level and using a 1–6 score scale (Aubertot et al., 2004). The disease severity scores were: 1 = healthy plant; 2 = less than 25% of the cross-sectioned stem girdled by lesion; 3 = 25%–50% of stem girdled by lesion; 4 = 50%–75% of stem girdled by lesion; 5 = 75%–90% of stem girdled by lesion, and 6 = 90%–100% of the cross-sectioned stem girdled by lesion. The mean disease severity (DS) was calculated using the following formula: $DS = (1 \times n) + (2 \times n) + (3 \times n) + (4 \times n) + (5 \times n) + (6 \times n) / N$, where n = number of plants within each specific score and N = total number of examined plants.

The incidence and severity of alternaria black spot was estimated at GS 85 on 30 stems and 100 pods per plot following the scale of Conn et al. (1990). The presence of sclerotinia stem rot was assessed on 30 plants and disease incidence (% of diseased plants) was also calculated.

Plant GSs were assessed according to Lancashire et al. (1991). The main meteorological data during the experimental period were taken from the Dotnuva hydrometeorological station, located about 1.5 km from the experimental fields (Table 1).

The seed yield from each plot was harvested separately by a small-plot harvester (Delta, Winterstieger, Austria). The seed moisture was estimated using a grain analyser (Infratec 1241, Foss, Denmark). Average seed yield ($t\ ha^{-1}$) of each cultivar in untreated and fungicide-treated plots was calculated and adjusted to 9% moisture content. TSW for seed samples was estimated using a seed-counter (Contador, Pfeuffer, Germany) and a balance (Explorer Ohaus, Ohaus Corporation, USA).

The data of disease incidence and severity, seed yield, and TSW were subjected to analysis of variance (ANOVA, split-plot) and tested for significance using the Fisher test. Treatment means were compared using least significant difference (LSD). The percentage data not following normal distribution were arcsine-transformed to stabilise variances.

3. Results

3.1. Phoma stem canker

The highest mean incidence of basal phoma canker (at GS 85) was obtained in 2008 and ranged in different cultivars from 61.2% to 75.0%, while the lowest was obtained in 2010, ranging from 35.8% to 52.5% (Figure 1). The effect of fungicides on the incidence of basal phoma canker varied among the growing seasons. Fungicide sprays exerted a very low and insignificant effect on the mean incidence of basal phoma canker in 2008. However, in 2009 and 2010, significant effect (F_{act}^{**} , $P < 0.001$ in 2009; F_{act}^* , $P < 0.001$ in 2010) of fungicide application was observed and mean incidence of basal canker was reduced significantly (from 58.4% to 49.4% in 2009, and from 58.7% to 33.3% in 2010). Mean incidence of basal phoma canker before harvest differed significantly among the cultivars in all 3 growing seasons (F_{act}^{**} , $P = 0.002$ in 2008; F_{act}^* , $P = 0.030$ in 2009; and F_{act}^* , $P = 0.045$ in 2010) (Table 2). Significantly ($P \leq 0.05$) lower incidence of basal canker as compared with Celsius was obtained for Siska and Titan in 2008. However, in 2009, significantly lower mean disease incidence was attributed to Casino, Insider, Siska, Sunday ($P \leq 0.01$), and Titan ($P \leq 0.05$). Cultivars Libea and Silvia were significantly ($P \leq 0.01$ and $P \leq 0.05$, respectively) less susceptible to basal phoma canker than Celsius in 2010.

As shown in Figure 2, the severity of basal phoma canker varied among the growing seasons; the highest mean disease severity in the untreated plots was achieved in 2008 (mean score 2.93), while the lowest was achieved in 2010 (mean score 1.94). With fungicide application we observed a significant decrease of the severity of basal canker during the 2009 and 2010 seasons (F_{act}^{**} , $P = 0.001$ in 2009; F_{act}^{**} , $P = 0.001$ in 2010) (Table 2). Some differences were also revealed among cultivars according to the mean severity of basal canker during all 3 growing seasons. However, significant effect of cultivar was obtained only in 2009 (F_{act}^{**} , $P = 0.003$). The data obtained from that season showed that the lowest mean value of the disease severity was estimated for Sunday and it was significantly lower in comparison with Celsius ($P \leq 0.01$). The mean

Table 1. Meteorological conditions during experimental period in May–July (Dotnuva hydrometeorological station).

Meteorological parameters	May			June			July		
	2008	2009	2010	2008	2009	2010	2008	2009	2010
Rainfall, mm	13.2	26.7	94.2	49.2	168.6	72.4	47.6	90.0	142.0
Relative air humidity, %	61.0	62.7	71.0	64.0	73.3	79.3	69.3	71.7	78.0
Days with rainfall of ≥ 1 mm	5	9	15	8	12	10	7	15	11
Mean air temperature, °C	12.2	12.7	13.7	16.1	14.6	16.2	18.2	18.1	21.7

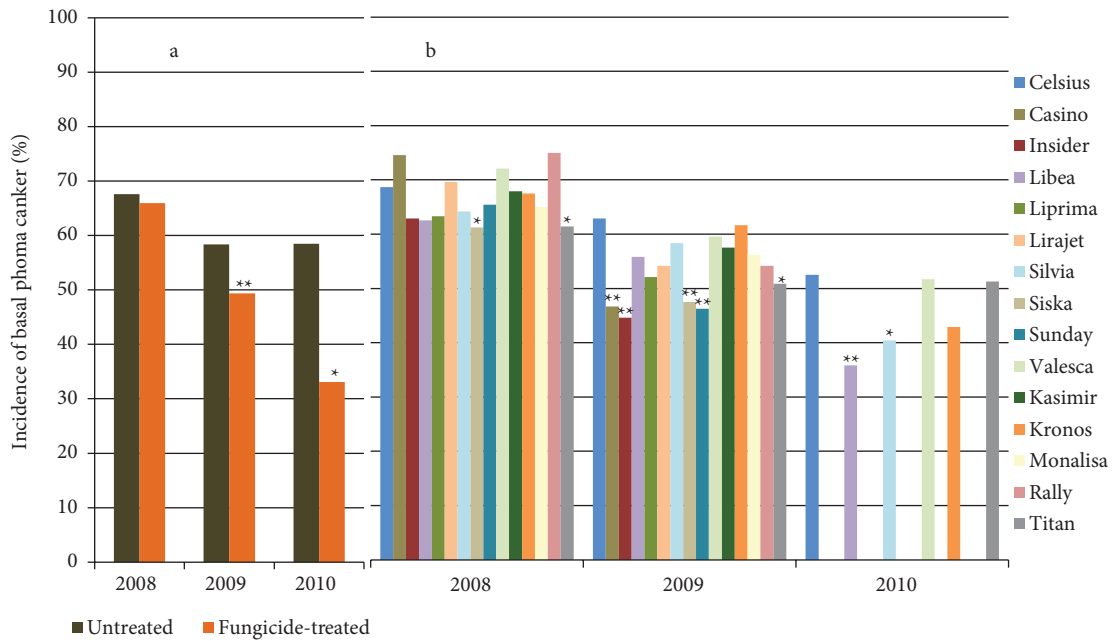


Figure 1. The effects of fungicide spray application (a) and winter rapeseed cultivar (b) on the incidence of basal phoma canker. *, **: significant differences from untreated plants (a) and cultivar Celsius (b) at P = 0.05 and P = 0.01, respectively.

Table 2. Analysis of variance P-values from fungal disease incidence and severity assessments, 2008–2010.

Source of variation	ANOVA values					
	2008		2009		2010	
	F _{act}	P	F _{act}	P	F _{act}	P
Incidence of basal phoma canker (%)						
Effect of fungicide application (A)	n.s.	0.245	**	<0.001	**	<0.001
Effect of cultivar (B)	**	0.002	*	0.030	*	0.045
A × B	n.s.	0.970	n.s.	0.508	n.s.	0.717
Significant treatment effect	n.s.	0.052	**	0.006	**	0.001
Severity of basal phoma canker (mean score)						
Effect of fungicide application (A)	n.s.	0.390	**	0.001	**	0.001
Effect of cultivar (B)	n.s.	0.367	**	0.003	n.s.	0.595
A × B	n.s.	0.999	n.s.	0.520	n.s.	0.364
Significant treatment effect	n.s.	0.927	**	0.004	*	0.021
Incidence of upper lesions of phoma stem canker (%)						
Effect of fungicide application (A)	n.s.	0.071	**	<0.001	**	0.004
Effect of cultivar (B)	**	<0.001	**	0.001	n.s.	0.271
A × B	n.s.	0.986	n.s.	0.804	n.s.	0.486
Significant treatment effect	**	<0.001	**	<0.001	n.s.	0.074
Severity of black spot on the stems (%)						
Effect of fungicide application (A)	**	<0.001	**	<0.001	**	<0.001
Effect of cultivar (B)	**	<0.001	**	0.001	n.s.	0.823
A × B	n.s.	0.142	*	0.020	n.s.	0.748
Significant treatment effect	**	<0.001	**	<0.001	**	<0.001
Severity of black spot on the pods (%)						
Effect of fungicide application (A)	**	0.001	**	<0.001	**	<0.001
Effect of cultivar (B)	*	0.049	**	<0.001	*	0.044
A × B	n.s.	0.412	*	0.016	n.s.	0.237
Significant treatment effect	**	0.009	**	<0.001	**	<0.001

*: P ≤ 0.05, **: P ≤ 0.01, n.s.: not significant.

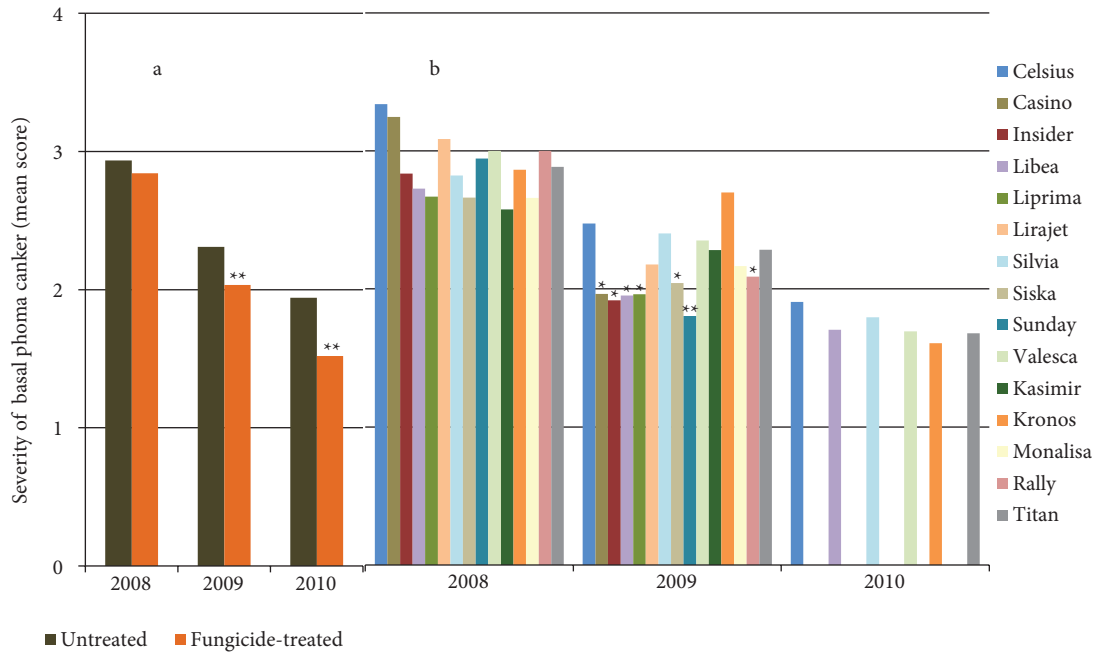


Figure 2. The effects of fungicide spray application (a) and winter rapeseed cultivar (b) on the severity of basal phoma canker. *, **: significant differences from untreated (a) and cultivar Celsius (b) at $P = 0.05$ and $P = 0.01$, respectively.

disease severity values estimated for Insider, Casino, Libea, Liprima, and Rally also differed significantly from that obtained for Celsius ($P \leq 0.05$).

The highest mean incidence of upper stem lesions was obtained in 2008 and varied among the cultivars from 47.5% to 86.0% (Figure 3). Significant effect of fungicide

spray application on the reduction of incidence of upper stem lesions was achieved during the 2009 (F_{act}^{**} , $P < 0.001$) and 2010 (F_{act}^{**} , $P = 0.004$) seasons. Significant effect of cultivar was obtained in 2008 and 2009 (F_{act}^{**} , $P < 0.001$ in 2008 and $P = 0.001$ in 2009, Table 2), and significantly ($P \leq 0.01$) lower disease incidence compared

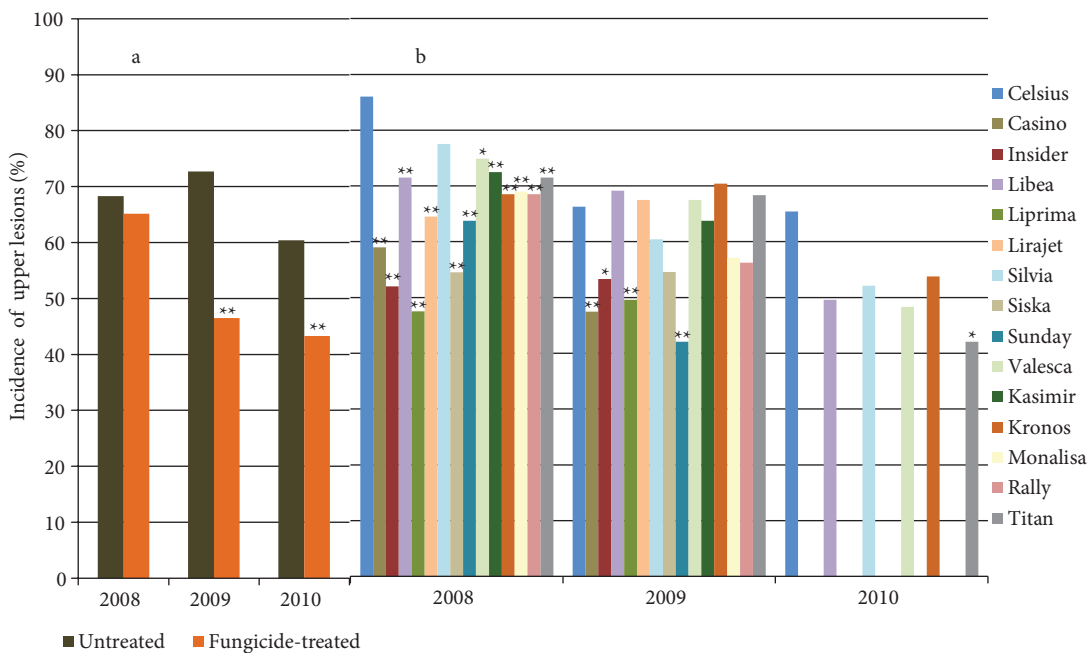


Figure 3. The effects of fungicide spray application (a) and winter rapeseed cultivar (b) on the incidence of upper lesions of phoma canker. *, **: significant differences from untreated (a) and cultivar Celsius (b) at $P = 0.05$ and $P = 0.01$, respectively.

with Celsius was revealed in almost all cultivars tested in 2008 (Figure 3). In 2009, however, significantly lower disease incidence was revealed only in Casino, Liprima, Sunday ($P \leq 0.01$), and Insider ($P \leq 0.05$).

3.2. Alternaria black spot and sclerotinia stem rot

As shown in Figures 4 and 5, the severity of alternaria black spot on the stems was higher in comparison with pods in 2008 and 2009. The data obtained in 2010, however, were controversial. Fungicide spray application significantly reduced disease severity on the stems and

pods in all 3 growing seasons (F_{act}^{**} , Table 2). Significant differences in susceptibility to black spot were estimated among the cultivars. The lowest disease severity on the stems during 2008 and 2009 was obtained in Sunday and Casino (significantly lower in comparison with Celsius, $P \leq 0.01$), as well as in Liprima and Siska ($P \leq 0.01$ in 2008 and $P \leq 0.05$ in 2009). Significantly lower disease severity on pods during 2008 and 2009 was obtained in Libea, Liprima, Silvia, and Rally in comparison with control cultivar Celsius.

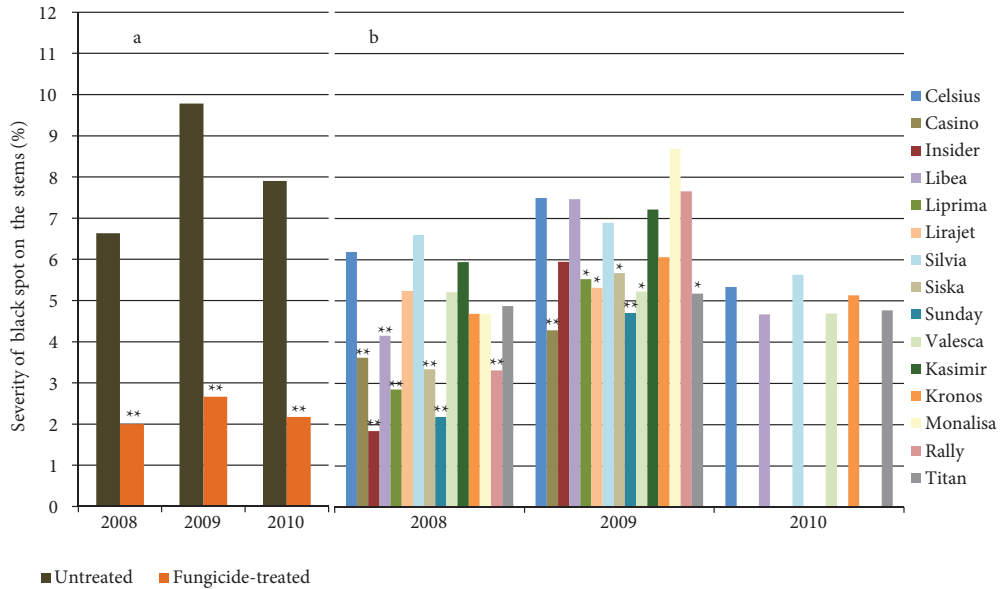


Figure 4. The effects of fungicide spray application (a) and winter rape cultivar (b) on the severity of alternaria black spot on the stems. *, **: significant differences from untreated (a) and cultivar Celsius (b) at $P = 0.05$ and $P = 0.01$, respectively.

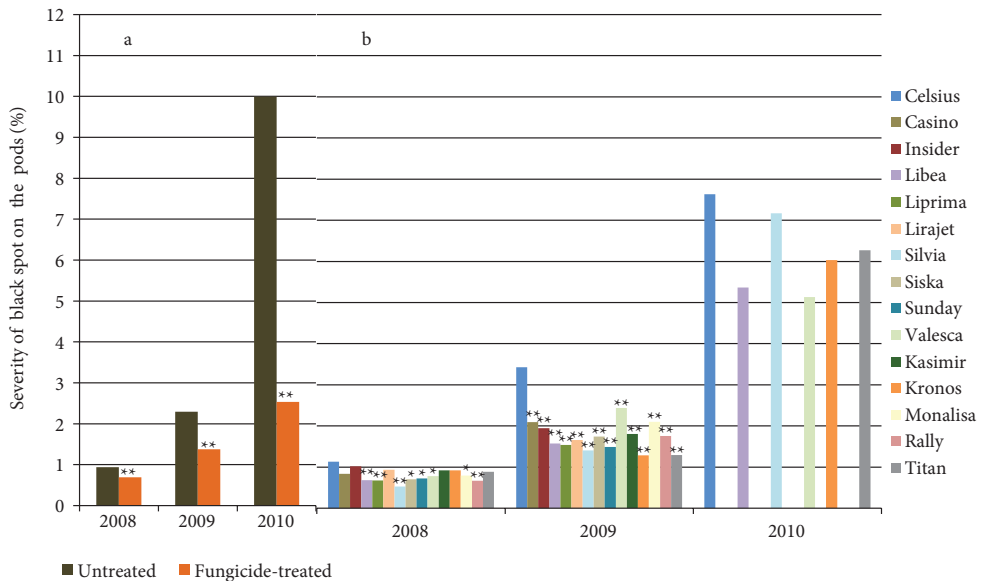


Figure 5. The effects of fungicide spray application (a) and winter rape cultivar (b) on the severity of alternaria black spot on the pods. *, **: significant differences from untreated (a) and cultivar Celsius (b) at $P = 0.05$ and $P = 0.01$, respectively.

Sclerotinia stem rot occurred in 2009 and 2010 on 6 cultivars tested and the disease incidence between the 2 seasons differed greatly (Table 3). Fungicide spray application completely controlled sclerotinia stem rot in 2009 (F_{act}^{**} , $P = 0.001$). In 2010, however, despite the fact that fungicide application significantly (F_{act}^{**} , $P < 0.001$) reduced disease incidence, an average 35.2% of plants in fungicide-treated plots were estimated to have symptoms of sclerotinia stem rot.

3.3. Seed yield, TSW, and economic benefit

During all 3 growing seasons, cultivar and fungicide application had a positive effect on the seed yield of winter oilseed rape (Table 4). In 2008, almost all cultivars tested produced significantly ($P \leq 0.01$) higher seed yields in comparison with Celsius and the highest mean seed yield was obtained in Rally, Monalisa, and Liprima (4.78, 4.54 and 4.50 t ha⁻¹, respectively). In 2009, the highest mean seed yield was produced by Sunday, Rally, Titan, Siska, Monalisa, Kronos, and Liprima (over 4.0 t ha⁻¹). The lowest seed yield was obtained in the 2009/10 growing season in comparison with the other 2 seasons. The cultivar Silvia produced a significantly lower seed yield in comparison with Celsius during all 3 seasons. The response of

cultivars to the fungicide application was variable during the experimental period. Significant and positive yield response to the fungicide application was obtained for all tested cultivars ($P \leq 0.01$) in 2009, and for 11 cultivars out of 15 tested in 2008 ($P \leq 0.01$). Yield increase due to fungicide application, however, was insignificant for all 6 cultivars tested in 2010.

The mean TSW was also affected by fungicide and cultivar (Table 5). Significant increase of TSW due to the fungicide application was obtained in all 3 growing seasons (F_{act}^{**} , $P < 0.001$). Positive TSW response of most cultivars to the fungicide application was obtained. The highest mean TSW during the experimental period was attributed to Insider (5.22 g), where it was significantly higher than that of the other tested cultivars. Significantly lowest mean TSW was recorded for Silvia and Siska.

Comparison of the economic benefit of the extra seed yield produced by the winter rape cultivars tested during 2 growing seasons showed additional income amounting to 130–334 euro ha⁻¹, and profit between 59–263 euro ha⁻¹ (Table 6). The profit from the cultivars tested for 3 growing seasons varied from 66 to 195 euro ha⁻¹.

Table 3. The effects of fungicide spray application at GS 63 on the incidence of sclerotinia stem rot on various cultivars of winter rape.

Effect of cultivar (B)	Effect of fungicide application (A)					
	2009			2010		
	Untreated	Treated	Mean (B)	Untreated	Treated	Mean (B)
Celsius	0.0	0.0	0.0	60.0	48.3	54.2
Libea	0.0	0.0	1.3	50.8	33.3	36.3**
Silvia	3.3	0.0	1.7*	70.0	47.5	41.1**
Valesca	1.7	0.0	0.0	53.3	30.0	42.1**
Kronos	3.3	0.0	1.7*	47.8	34.4	58.7*
Titan	2.5	0.0	0.8	55.0	17.5	41.7**
Mean (A)	1.8	0.0**		56.2	35.2**	
Analysis of variance						
	F_{act}	P-values		F_{act}	P-values	
A	**	0.001		**	<0.001	
B	n.s.	0.060		**	<0.001	
A × B	n.s.	0.060		**	0.001	
Significant treatment effect	**	0.001		**	<0.001	

*: $P \leq 0.05$, **: $P \leq 0.01$, n.s.: not significant.

Table 4. The effects of fungicide spray application and cultivar on the seed yield (t ha⁻¹) of winter rape.

Effect of cultivar (B)	Effect of fungicide application (A)								
	2008			2009			2010		
	Untreated	Treated	Mean (B)	Untreated	Treated	Mean (B)	Untreated	Treated	Mean (B)
Celsius	3.07	3.55	3.31	3.44	3.96	3.70	2.85	3.12	2.98
Casino	3.14	3.94	3.54*	3.53	4.18	3.86	-	-	-
Insider	4.20	4.24	4.22**	3.64	4.37	4.00*	-	-	-
Libea	3.59	4.12	3.85**	3.64	4.25	3.94*	2.61	2.91	2.76
Liprima	4.38	4.62	4.50**	3.71	4.51	4.11**	-	-	-
Lirajet	3.17	3.96	3.56*	3.33	4.21	3.77	-	-	-
Silvia	2.05	2.61	2.33**	2.85	3.32	3.08**	1.96	2.04	2.00**
Siska	3.98	4.27	4.12**	3.82	4.73	4.27**	-	-	-
Sunday	4.27	4.28	4.28**	4.09	4.73	4.41**	-	-	-
Valesca	2.46	3.63	3.04**	3.48	4.06	3.77	2.47	2.72	2.59
Kasimir	3.61	3.95	3.78**	3.62	4.19	3.90	-	-	-
Kronos	3.93	4.45	4.19**	3.87	4.50	4.18**	2.85	3.02	2.93
Monalisa	4.52	4.55	4.54**	3.92	4.60	4.26**	-	-	-
Rally	4.58	4.98	4.78**	4.14	4.51	4.32**	-	-	-
Titan	4.33	4.40	4.37**	3.91	4.69	4.30**	3.00	3.18	3.09
Mean (A)	3.68	4.10**		3.66	4.32**		2.62	2.83	

Analysis of variance						
	F _{act}	P-values	F _{act}	P-values	F _{act}	P-values
A	**	<0.001	**	<0.001	n.s.	0.168
B	**	<0.001	**	<0.001	**	0.002
A × B	**	0.001	n.s.	0.707	n.s.	0.707
Significant treatment effect	**	<0.001	**	<0.001	*	0.023

*: $P \leq 0.05$, **: $P \leq 0.01$, n.s.: not significant.

4. Discussion

In recent years, intensive cultivation of winter and spring oilseed rape in Lithuania has increased the occurrence of numerous fungal diseases. Effective control of phoma stem canker can be achieved through the choice of resistant cultivars; however, for the control of alternaria black spot and sclerotinia stem rot only fungicide spray applications can be helpful. Moreover, foliar fungicides can also give good control of stem canker and result in significant increase in seed yield of winter rape.

Infection of phoma stem canker in winter rape starts in the autumn from ascospores, which are produced and discharged from pseudothecia on rape stubble (McGee and Petrie, 1979). Ascospores infect leaves and first phoma leaf spots develop, then further infection from leaf spots grows through petioles and finally reaches stems, where it becomes necrotrophic and causes basal canker with visual symptoms before harvest (Hammond et al., 1985; Fitt et al., 2006b). Chemical control of phoma basal canker is possible only by fungicide application in the autumn.

Table 5. The effects of fungicide spray application and cultivar on the thousand-seed weight (g) of winter rape.

Effect of cultivar (B)	Effect of fungicide application (A)								
	2008			2009			2010		
	Untreated	Treated	Mean (B)	Untreated	Treated	Mean (B)	Untreated	Treated	Mean (B)
Celsius	4.54	4.68	4.61	4.74	5.23	4.99**	5.32	5.79	5.55
Casino	4.64	4.82	4.73	4.62	5.08	4.85*	-	-	-
Insider	5.14	5.32	5.22**	5.00	5.44	5.22**	-	-	-
Libea	4.36	4.61	4.49	4.40	4.73	4.56**	5.11	5.19	5.15**
Liprima	4.93	5.00	4.96**	4.59	4.94	4.76**	-	-	-
Lirajet	4.30	4.52	4.41*	4.13	4.59	4.36**	-	-	-
Silvia	3.96	3.97	3.97**	3.86	4.14	4.00**	4.57	4.68	4.62**
Siska	3.96	4.18	4.07**	3.95	4.26	4.11**	-	-	-
Sunday	4.31	4.47	4.39**	4.51	4.92	4.72**	-	-	-
Valesca	4.09	4.40	4.25**	4.26	4.74	4.50**	4.83	5.20	5.01**
Kasimir	4.24	4.47	4.36**	4.39	4.79	4.59**	-	-	-
Kronos	4.33	4.57	4.45*	4.22	4.58	4.40**	5.45	5.95	5.70**
Monalisa	4.35	4.44	4.39**	4.34	4.73	4.54**	-	-	-
Rally	4.69	4.89	4.79*	4.37	4.76	4.56**	-	-	-
Titan	4.46	4.66	4.56	4.14	4.63	4.38**	5.11	5.71	5.41**
Mean (A)	4.42	4.60**		4.37	4.77**		5.06	5.42**	

Analysis of variance						
	F _{act}	P-values	F _{act}	P-values	F _{act}	P-values
A	**	<0.001	**	<0.001	**	<0.001
B	**	<0.001	**	<0.001	**	<0.001
A × B	n.s.	0.928	n.s.	0.779	**	<0.001
Significant treatment effect	**	<0.001	**	<0.001	**	<0.001

*: $P \leq 0.05$, **: $P \leq 0.01$, n.s.: not significant.

Disease incidence on winter rape leaves in autumn (usually 10%–20% of plants with phoma leaf spots) is an indicator for fungicide application against phoma stem canker (West et al., 2002; Gladders et al., 2006; Steed et al., 2007). Despite the fact that during our experimental period fungicide was applied in autumn at the optimal time (when the incidence of phoma leaf spot ranged from 10% to 25%), the effect of the autumn fungicide spray application on the incidence and severity of basal stem canker was not always meaningful. For example,

fungicide spray application was not effective during the 2007/08 cropping season, when the disease incidence and severity were the highest during our experimental period. Previous reports also show that applying fungicide sprays sometimes provided little control of phoma disease (Fitt et al., 1997; Gladders et al., 1998). However, significant effect of fungicide application on phoma basal canker was achieved during the second and third experimental years and this is in agreement with numerous reports, suggesting that foliar fungicides give good control of basal

Table 6. Mean profitability of double fungicide application (metconazole at GS 15–16 and boscalid at GS 63) in various cultivars of winter rape.

Cultivar	Mean of 2 growing seasons			Cultivar	Mean of 3 growing seasons		
	Extra yield (kg ha ⁻¹)	Additional income (euro ha ⁻¹)	Profit (euro ha ⁻¹)		Extra yield (kg ha ⁻¹)	Additional income (euro ha ⁻¹)	Profit (euro ha ⁻¹)
Casino	725	290	219	Celsius	425	170	99
Insider	386	154	83	Libea	481	192	121
Liprima	521	208	137	Silvia	370	148	77
Lirajet	835	334	263	Valesca	666	266	195
Siska	602	241	170	Kronos	443	177	106
Sunday	325	130	59	Titan	342	137	66
Kasimir	454	182	111	Mean	455	182	111
Monalisa	355	142	71				
Rally	381	152	81				
Mean	509	204	133				

Price of rape seed yield was 400 euro t⁻¹.

Prices of fungicides Juventus (metconazole) and Cantus (boscalid) were 27 and 78 euro L⁻¹, respectively.

The cost of fungicide application was 6 euro ha⁻¹.

The cost of double fungicide application (including fungicides) was 71 euro ha⁻¹.

cankers (West et al., 1999; Zhou et al., 1999; Gladders et al., 2004a, 2004b). Variable efficacy of fungicide application on the incidence and severity of basal phoma canker was most likely influenced by the differences in the conditions for the phoma infection during the experimental period. It was stated by Marcroft et al. (2004) that environmental conditions may considerably influence phoma infection. The benefits from foliar fungicide treatments are ambiguous due to the long periods of dissemination of ascospores of the *L. maculans* and *L. biglobosa* species complex and relatively short periods of fungicide persistence. Efficacy of triazole fungicide metconazole was likely to have also been influenced by the proportions of *L. maculans* and *L. biglobosa*, the 2 coexisting *Leptosphaeria* species on oilseed rape in Lithuania (Brazauskienė et al., 2011b). It was shown by Eckert et al. (2010) and Huang et al. (2011) that triazole fungicides flusilazole and tebuconazole were more effective against *L. maculans* than against *L. biglobosa*.

Winter rape cultivars showed significant differences in their susceptibility to basal phoma canker and upper stem lesions; however, neither of the cultivars showed high resistance to this disease. Amongst the cultivars tested, the lowest disease incidence and severity were revealed in Casino, Insider, Sunday, Siska, and Titan. Unfortunately, there was no significant cultivar × treatment interaction.

The differences in the incidence and severity of other fungal diseases during the experimental period could be explained by the differences in the meteorological conditions, especially the amount and frequency of precipitation. In 2008, fungicide application significantly reduced the severity of alternaria black spot on the stems; however, due to the dry conditions unfavourable for black spot infection during June and July, the disease severity on the pods was very low. Meteorological conditions were conducive to fungal diseases in 2010 and high pressure of alternaria black pod spot and sclerotinia stem rot were recorded during that season. This is in agreement with other reports that meteorological conditions can greatly affect the spread and severity of alternaria black spot (Hong et al., 1996; Shrestha et al., 2005) and sclerotinia stem rot (Mila et al., 2003; Kutcher and Wolf, 2006). During our research, fungicide application significantly reduced the severity of alternaria black spot on stems and pods. Despite the fact that the cultivars differed in their susceptibility to alternaria black spot, our findings are in agreement with those of other authors, reporting that in the absence of stable sources of resistance to *A. brassicae*, chemical fungicides are the only effective measure to manage alternaria black spot (Chattopadhyay and Bagchi, 1994). During our research, fungicide application also

significantly reduced the incidence of sclerotinia stem rot; however, its efficacy during the season with high disease pressure was insufficient. As was suggested by other researchers, control of the diseases caused by *S. sclerotiorum* is complicated (Bardin and Huang, 2001; Ma et al., 2009).

During our research, fungicide spray applications increased the seed yield and TSW of winter rape; however, significant seed yield increase was obtained only in 2008 and 2009. As was discussed above, control of phoma stem canker in 2008 was not very effective; only alternaria black spot was controlled significantly. Since alternaria black spot severity on pods during that season was very low, it seems that significant yield increase was obtained due to effective control of alternaria black spot on the stems; this also might be due to some physiological effect of fungicide application. In 2009, seed yield increase was obtained due to effective control of phoma stem canker (basal canker and upper stem lesions), alternaria black spot on stems and pods, and sclerotinia stem rot. This is in agreement with other reports that foliar fungicides have given good control of diseases and resulted in significant increases in yield of oilseed rape (Gladders et al., 1998; West et al., 1999; Zhou et al., 1999; Balodis et al., 2008). On the other hand, Gladders et al. (2006) noticed that economic efficacy of fungicide use depends on yield benefits in relation to disease severity (dependent on seasonal conditions) and host resistance. Our results support these findings. Fungicide use in 2010

gave effective control of fungal diseases; however, seed yield increase was not significant, which was probably due to insufficient control of sclerotinia stem rot during that season.

Huang et al. (2011) reported that cultivars differ in their yield response to the fungicide treatment. During our research, significant and positive yield response to the fungicide application was obtained in almost all tested cultivars in 2008 and 2009; however, the cultivars that were higher yielding without treatment (Insider, Sunday, Monalisa, Rally, and Titan) showed lower response to the fungicide application in 2008 but not in 2009. The lowest seed yield of Silvia in all 3 growing seasons can be explained by the very low TSW of that cultivar.

In general, application of fungicides was profitable during the experimental period for all cultivars tested. During our research, the use of fungicides demonstrated the highest economic return from the cultivars Lirajet, Casino, Siska, Liprima, and Valesca. In Lithuania, the price of rape seed yield has been variable (200–450 euro t⁻¹) in recent years; therefore, profitability of fungicide spray application can vary between cropping seasons.

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