

## THE EFFECT OF ADJUVANTS, SPRAY VOLUME AND NOZZLE TYPE ON AZOXYSTROBIN EFFICACY AGAINST *LEPTOSPHAERIA MACULANS* AND *L. BIGLOBOSA* ON WINTER OILSEED RAPE

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**Abstract:** In two vegetative seasons 2005–2006 azoxystrobin (Amistar 250 SC at the dose of 0.7 dm<sup>3</sup>/ha) was applied as a spraying treatment at the beginning of plant flowering against stem canker (*Leptosphaeria maculans* and *L. biglobosa*) in winter oilseed rape crops. The effect of the following spray application parameters: water volume (200 and 400 l/ha), adjuvant type (Break Thru S 240 – 0.1% and Atpolan 80 EC – 0.5%), and nozzle type (XR11002 – fine droplet size and DB11002 – coarse droplets at 0.4 MPa pressure) on the fungicide efficacy was studied. The results of presented studies indicated that azoxystrobin (Amistar 250 SC) caused a significant decrease of rape infection by stem canker. Addition of adjuvants did not significantly increase the fungicide effectiveness, and sometimes lowered its action (especially after the application of Atpolan 80 EC). The applied spray volumes of water and nozzle type provided an effective protection of oilseed rape against stem canker, however, in some cases the results of field experiments were not significantly different. The best results of the disease casual agent (*L. biglobosa*) control, statistically proved were shown at the higher spray volume (400 l/ha), regardless of nozzle type (droplet size) and adjuvant application.

**Key words:** azoxystrobin, stem canker, *Leptosphaeria maculans*, *L. biglobosa*, nozzle type, spray volume, adjuvant

### INTRODUCTION

Stem canker caused by two pathogens – *Leptosphaeria maculans* and *L. biglobosa* – is considered as a worldwide disease of oilseed rape, including Poland (Fitt *et al.* 2006; Jędrzycka 2006). Among methods for the control of the disease casual agent, spraying with fungicides is the most important treatment. According to the recommendations worked out by the Institute of Plant Protection – National Research Institute in Poznań two terms of fungicide application are possible (Gwiazdowski 2002). The first date when spraying should be carried out is the autumn, at the growth stage of 4 leaves before the rosette appearing, or after the first disease symptoms are observed. The second spraying is possible in spring after start of plant vegetation or when first symptoms are noticed.

The spring application of fungicides might be considered as problematic due to a possible occurrence of plants already infected in autumn and in early spring. Pathogen airborne ascospores are released for several months – from September to May (Jędrzycka 2006). Winter oilseed rape plants are usually high in spring, so it may be difficult to spray precisely the lowest stem parts and leaves

near soil surface. However, the spring treatment allows the active ingredient to remain in sprayed plants and protect them against later infections, caused mainly by *L. biglobosa* (Sadowski and Klepin 1991; Gwiazdowski 2002).

Several active ingredients can be used for the control of stem canker. The majority of them belong to the groups of triazoles and benzimidazoles, systemic fungicides, which can be transported inside plants. It should be emphasized that the efficacy of spray application depends on plant precise covering by chemicals, retention, deposition and ability of fungicide penetration into plant tissues. Those features can be modified by addition of adjuvants, adequate spray volume and a proper droplet size (Gaskin *et al.* 2000; Holloway *et al.* 2000).

The aim of presented studies was to evaluate the efficacy of azoxystrobin (Amistar 250 SC) in the control of stem canker (*L. maculans* and *L. biglobosa*) in winter oilseed rape crops by a combined action of adjuvants, volume of liquid per hectare and proper size of spray droplets.

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## MATERIALS AND METHODS

The experiment was carried out in the province of Wielkopolska on winter oilseed rape in two seasons: 2005–2006 in Skórzewo and 2006–2007 in Złotniki, using cv. Lisek. Field experiments were established using randomized block design in 4 replications with the plot size of 2x10 m. The previous crop was winter oilseed rape grown in both years, to increase the infection pressure of *Leptosphaeria* sp. The fungicide Amistar 250 SC was applied using knapsack sprayer at the dose of 0.7 dm<sup>3</sup>/ha at BBCH 59–61 (beginning of oilseed rape flowering). A spray boom was established 25–30 cm above plant canopy. In both years, wind speed during performed treatments did not exceed 2 m/s and the temperature 25°C (tab. 1). The height of plants differed between the

years. In 2005, the mean height was 71 cm but in 2006 it was much higher and amounted to 130 cm. The effect of 3 factors: 1) volume of water (200 and 400 dm<sup>3</sup>/ha), 2) adjuvant type (Break Thru S 240 in 0,1% concentration and Atpolan 80 EC in 0,5% concentration), and 3) nozzle type (XR11002 – fine droplet size and DB11002 – coarse droplets at 0,4 MPa pressure) were studied. The control treatments were untreated plots. Control of weeds, pests and other agrotechnical treatments were carried out according to respective recommendations in a homogeneous way on the whole plantation.

The evaluation of infection caused by the pathogens was carried out on 25 plants collected from each plot. Leaf infection was recorded using a 5 degree scale: 0 – healthy; 1 – 1 to 2 spots on leaf surface; 2 – 3 to 4; 3 – 5 to 6; 4 – 7 or more spots. For the assessment of stem infection by

Table 1. The meteorological conditions during field experiments

Month	Decade	2005 (Skórzewo)			2006 (Złotniki)		
		sum	mean		sum	mean	
		rainfall [mm]	temperature [°C]	relative humidity [%]	rainfall [mm]	temperature [°C]	relative humidity [%]
April	I	9	9.29	65.15	0.6	7.32	77.40
	II	0	11.15	74.47	6	9.04	77.89
	III	7.4	8.35	61.67	23.4	11.47	84.44
	sum/mean	16.40	9.59	67.10	30.00	9.28	79.91
May	I	29.2	11.49	86.04	11.6	14.80	61.00
	II	21.8	10.10	79.77	13.6	14.87	75.33
	III	11.2	19.20	67.51	20.4	12.59	80.73
	sum/mean	62.20	13.59	77.77	45.60	14.09	72.35
June	I	8.6	13.33	75.85	6.8	12.80	80.36
	II	8	16.94	73.02	0.4	20.83	67.91
	III	0.2	19.71	63.28	14.4	21.92	64.60
	sum/mean	16.80	16.66	70.72	21.60	18.52	70.96
July	I	9.4	19.98	69.51	0.0	22.44	44.02
	II	3.6	21.08	63.07	20.2	22.73	62.80
Sum/Mean		108.40	14.60	70.85	117.40	15.53	70.59

*L. maculans* and *L. biglobosa*, a 6 degree scale was used (Aubertot *et al.* 2004; Karolewski *et al.* 2009), where 1 meant healthy plant and 6 plant infected at 90–100%.

Plant samples were collected once in 2005, in pre ripening phase (BBCH 87 – stem infection), while in 2006, three assessments were performed: at the BBCH 67–68 (25.05 – leaf infection), 77 (27.06 – stem infection) and 87 (17.07 – stem infection).

The results of field experiments were subjected to statistical analysis using Duncan's test at significance level  $\alpha = 0.05$ .

## RESULTS

The level of stem infection before harvesting caused by the fungi *L. maculans* and *L. biglobosa* (untreated plots) was slightly higher in 2005 comparing to 2006 (Table 2, 3).

Table 2. The effect of Amistar 250 SC spraying [0.7 dm<sup>3</sup>/ha] on winter oilseed rape infection, internal stem symptoms (*L. maculans*) depending on spray volume, adjuvant and nozzle type

Spray volume [dm <sup>3</sup> /ha]	Adjuvant	Nozzle type	2005		2006						
			stems before harvest		leaves (BBCH-67-68)		stems (BBCH 77)		stems before harvest		
				mean	25.05	mean	27.06	mean		mean	
200	-	XR 11002	1.44* a	1.42 a	0.29* a	0.32 a	1.44 a	1.50 a	1.72 a	1.65 a	
		DB 12002	1.40* a		0.35* a		1.55 a		1.58 a		
	Atpolan 80 EC	XR 11002	1.77 a	1.71 b	0.32* a	0.29 a	1.61 a	1.57 a	1.60 a	1.59 a	
		DB 12002	1.64* a		0.26* a		1.52 a		1.58 a		
	Break Thru S 240	XR 11002	1.53* a	1.53 ab	0.27* a	0.24 a	1.51 a	1.62 a	1.58 a	1.59 a	
	Mean	DB 12002	1.52* a	1.55 A	0.20* a	0.28 A	1.73 a	1.56 A	1.60 a	1.61 A	
	400	-	XR 11002	1.53* a	1.52 ab	0.37* a	0.33 a	1.49 a	1.44 a	1.65 a	1.62 a
			DB 12002	1.50* a		0.29* a		1.39 a		1.59 a	
		Atpolan 80 EC	XR 11002	1.61* a	1.59 ab	0.28* a	0.33 a	1.73 a	1.55 a	1.70 a	1.45 a
DB 12002			1.57* a	0.37* a		1.36 a		1.54 a			
Break Thru S 240		XR 11002	1.42* a	1.46 ab	0.20* a	0.23 a	1.51 a	1.58 a	1.45 a	1.62 a	
		DB 12002	1.49* a		0.26* a		1.65 a		1.45 a		
Mean			1.52 A		0.30 A		1.52 A		1.56 A		
Control (untreated plots)			2.07		0.90		1.40		1.67		
Mean		-	XR 11002	1.49 a			0.33 a		1.47 a		1.69 a
			DB 12002	1.45 a			0.32 a		1.47 a		1.65 a
	Atpolan 80 EC	XR 11002	1.69 a			0.30 a		1.67 a		1.65 a	
		DB 12002	1.61 a			0.32 a		1.44 a		1.59 a	
	Break Thru S 240	XR 11002	1.48 a			0.24 a		1.51 a		1.52 a	
		DB 12002	1.51 a			0.23 a		1.69 a		1.59 a	

a, b or A –the same letter indicate significant differences according to Duncan test at  $\alpha = 0.05$

\* mean significantly different from control (untreated) by Duncan test at  $\alpha = 0.05$

The level of leaf infection (based on the results of observations in 2006) caused by *L. maculans* was higher than the one caused by *L. biglobosa*.

Application of azoxystrobin (Amistar 250 SC) caused a significant decrease of rape infestation by both pathogens, with the exception of some observation dates and experimental treatments where no significant differences were recorded. Adjuvants and parameters of spraying treatment considerably contributed to the level of oilseed rape infection especially in case of *L. biglobosa* (Table 2, 3).

Water volume used for the dilution of fungicide without any addition of adjuvant did not affect significantly the level of rape infestation by stem canker. However, after the addition of adjuvant, a slightly less intensive infec-

tion of rape leaves was recorded especially with the use of a higher water volume (with the exception of rape leaf infection caused by *L. maculans*) (Table 2, 3).

Finally, it was shown that higher spray volume provided a better protection of rape against *L. biglobosa*, as it was revealed before the harvest in 2006.

Adjuvants did not affect significantly the efficacy of azoxystrobin. However, in 2005, the addition of Atpolan 80 EC to spray volume of 200 l/ha, resulted in a less effective rape protection against *L. biglobosa* and *L. maculans* comparing to the treatments without adjuvant.

The type of nozzle used for the liquid application did not exert any significant effect on the obtained results of rape protection against stem canker, with the exception

Table 3. The effect of Amistar 250 SC spraying (0.7 dm<sup>3</sup>/ha) on winter oilseed rape stem infection, external stem symptoms (*L. biglobosa*) depending on spray volume, adjuvant and nozzle type

Spray volume [dm <sup>3</sup> /ha]	Adjuvant	Nozzle type	2005		2006						
			stems before harvest		leaves (BBCH-67-68)		stems (BBCH 77)		stems before harvest		
				mean	25.05	mean	27.06	mean		mean	
200	-	XR 11002	1.60 ab	1.52 a	0.26 ab	0.24 a	1.49 a	1.53 a	1.55 a	1.54 a	
		DB 12002	1.44* a		0.22 ab		1.56 a		1.52 a		
	Atpolan 80 EC	XR 11002	1.86 b	1.79 b	0.34 b	0.29 a	1.7 a	1.53 a	1.60 a	1.52 a	
		DB 12002	1.72 ab		0.23 ab		1.35 a		1.43* a		
	Break Thru S 240	XR 11002	1.44* a	1.49 a	0.32 ab	0.28 a	1.61 a	1.62 a	1.52 a	1.52 a	
		DB 12002	1.53 a	1.6 A	0.23 ab	0.27 A	1.62 a	1.55 A	1.52 a	1.52 B	
	Mean										
	400	-	XR 11002	1.50 a	1.46 a	0.25 ab	0.25 a	1.58 a	1.56 a	1.40* a	1.41 a
			DB 12002	1.43* a		0.25 ab		1.54 a		1.42* a	
		Atpolan 80 EC	XR 11002	1.62 ab	1.65 ab	0.28 ab	0.21 a	1.48 a	1.45 a	1.56 a	1.46 a
DB 12002			1.68 ab	0.15 a		1.41 a		1.36* a			
Break Thru S 240		XR 11002	1.40* a	1.43 a	0.20 ab	0.22 a	1.44 a	1.50 a	1.35* a	1.41 a	
		DB 12002	1.45* a		0.21 ab		1.55 a		1.46* a		
Mean			1.51 A		0.22 A		1.50 A		1.43 A		
Control (Untreated plots)			1.82		0.29		1.52		1.77		
Mean		-	XR 11002	1.55 abc		0.26 a		1.54 a		1.48 a	
			DB 12002	1.43 a		0.24 a		1.55 a		1.47 a	
	Atpolan 80 EC	XR 11002	1.74 c		0.31 a		1.59 a		1.58 a		
		DB 12002	1.70 bc		0.19 a		1.38 a		1.40 a		
	Break Thru S 240	XR 11002	1.42 a		0.26 a		1.53 a		1.44 a		
		DB 12002	1.49 ab		0.22 a		1.59 a		1.49 a		

a, b or A – the same letter indicate significant differences according to Duncan test at  $\alpha = 0.05$

\* mean significantly different from control (untreated) by Duncan test at  $\alpha = 0.05$

of *L. biglobosa* on leaves when the interaction with different spray volumes was analysed. Air induction nozzle (DB 12002) used for the application of Amistar 250 SC with Atpolan 80 EC and 400 l/ha was shown to be better than the standard nozzle (XR 11002) with spray volume of 200 l/ha.

## DISCUSSION

One spring chemical treatment to protect oilseed rape against stem canker seems to be effective enough (Gwiazdowski *et al.* 2002), in spite of a possible occurrence of disease casual agent *L. maculans* in autumn (Fitt *et al.* 2006; Jedryczka 2006), and spring infection caused by

*L. maculans* and *L. biglobosa* which may last for a long time after performed spraying treatments. A high intensity of infection is usually observed in May and June, as a result of favorable thermal conditions, and the amount of rainfall and its distribution. In May 2005, rainfall was 36% higher comparing to 2006, and probably affected the increase of *Leptosphaeria* spp. infection. In 2005 during the last assessment of stems, the degree of infection caused by *L. maculans* on untreated plants reached 2.07, while in 2007, it was only 1.67. The degree of infection caused by *L. biglobosa* reached only 1.22 in 2005 and 1.77 in 2007. The method of disease symptom evaluation which was used in presented studies only included internal stem symptoms in case of *L. maculans* and external stem symp-

toms of infection caused by *L. biglobosa*, however internal symptoms can be also caused by *L. biglobosa* (Eckert *et al.* 2004; Jędrzycka 2006).

Azoxystrobin showed a good efficacy in oilseed rape protection against both pathogens, in spite of the fact that this fungicide was not recommended in Poland. The results of azoxystrobin application were similar to the ones obtained with the use of metconazole in the same period (Karolewski *et al.* 2009). More frequent significant differences in the efficacy of azoxystrobin in case of *L. biglobosa* were probably due to the parts of oilseed rape plants infected by the pathogen.

Water volume used for the dilution of fungicides may affect their efficacy. The range of water volume accepted for these studies, both 200 dm<sup>3</sup> and 400 dm<sup>3</sup> provided an effective action of azoxystrobin. It is known that the dosage of water volume depends largely on the height and size of plants. In spite of the fact that during the treatment, the plants were much higher in 2006 than in 2005, only in case of *L. biglobosa* (evaluation before harvest), a better efficiency was achieved in 2006 by applying a higher water volume, regardless of the presence of adjuvants and different nozzle type. Therefore, the spray volume of 200 dm<sup>3</sup> seems to be sufficient enough to provide a good azoxystrobin action on rape plants with an average height not exceeding 71 cm. Karolewski *et al.* (2009) reported that spray volume of 200 dm<sup>3</sup> also provided a good effectiveness of metconazole application against stem canker.

The results of performed studies did not clearly reveal a positive effect of adjuvants on the action of azoxystrobin and even they lowered the efficiency of applied treatments especially in case Atpolan 80 EC. The positive effect of adjuvants on the fungicide efficacy was more visible when it was applied at a small dose (Ratajkiewicz *et al.* 2008). This might explain why no significant differences in rape protection were obtained either with the use of azoxystrobin or metconazole (Karolewski *et al.* 2009). Ratajkiewicz *et al.* (2008) reported that the efficiency of tetraconazole against cercospora leaf spot of beets was significantly higher with the use of the adjuvant Break Thru S 240 comparing to the fungicide applied alone.

Grayson *et al.* (1996) and Kierzek and Wachowiak (2003) suggested that an addition of adjuvants might not improve the efficacy of fungicide spraying without taking into consideration other application parameters. Appropriate selection of spray application parameters such as nozzle type, water volume, adjuvant type may effectively improve a fungicide action. Kutcher and Wolf (2006) reported that the use of volume are appropriate for the control of Brassica stem diseases. The results of performed studies did not confirm those reports. However, it seems justified that air induction nozzle DB 11002 producing coarse droplet size can be recommended in winter oilseed rape crops, especially if the spraying is performed at high speed wind (3–5 m/s). Finally, one can state that both nozzles used in our studies are useful for rape protection against stem canker.

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## POLISH SUMMARY

### WPŁYW ADJUWANTÓW, OBJĘTOŚCI CIECZY I TYPU ROZPYLACZA NA SKUTECZNOŚĆ DZIAŁANIA AZOKSYSTROBINY W OCHRONIE RZEPAKU PRZED *LEPTOSPHAERIA MACULANS* I *L. BIGLOBOSA*

W latach 2005 i 2006 na początku kwitnienia rzepaku ozimego zastosowano azoxystrobinę (Amistar 250 SC w dawce 0,7 dm<sup>3</sup>/ha) przeciwko suchej zgniliznie ka-

pustnych (*Leptosphaeria maculans* i *L. biglobosa*). Oceniono znaczenie ilości wody (200 i 400 l/ha), rodzaju adiuwanta (Break Thru S 240 – 0,1% i Atpolan 80 EC – 0,5%) oraz rodzaju rozpylacza (XR 11002 – drobnokroplisty i DB11002 – grubokroplisty przy ciśnieniu 0,4 MPa), dla biologicznej skuteczności opryskiwania fungicydem.

Wyniki badań wskazują, że azoxystrobina (Amistar 250 SC) przyczyniła się do istotnego obniżenia porażenia rzepaku przez suchą zgniliznę, jednak dodatek adiuwantów nie wpływał istotnie na wzrost skuteczności jej dzia-

łania, a nawet niekiedy – po zastosowaniu Atpolan 80 EC – był powodem jej obniżenia. Zastosowane ilości wody i rozpylacze pozwoliły na skuteczną ochronę rzepaku przed suchą zgnilizną, a uzyskane wyniki zazwyczaj nie różniły się istotnie. Tylko *L. biglobosa* w ocenie przed zbiorem w 2006 roku była skuteczniej zwalczana w kombinacjach z użyciem większej dawki wody (400 l/ha), bez względu na rodzaj zastosowanego rozpylacza (wielkość kropli) i dodatku adiuwanta.