P L A N T B R E E D I N G A N D S E E D S C I E N C E
Volume 56 2007

Irena Kiecana, Małgorzata Cegiełko

Department of Phythopatology, Agricultural University of Lublin,Leszczyńskiego 7, 20-069 Lublin, Poland e-mail: irena.kiecana@ar.lublin.pl

PATHOGENICITY OF *BIPOLARIS SOROKINIANA* (SACC.) SHOEM. TO SELECTED OAT (*AVENA SATIVA* L.) GENOTYPES

ABSTRACT

Field experiments with artificial inoculation of soil and grain of 12 oat genotypes with *Bipolaris sorokiniana* isolate No. 36 were carried out in years 2000-2002 in Zamość region (south-eastern part of Poland). In each year the number of 7-weeks seedlings, number of plants and panicles before harvest and kernels yield from the individual plot were calculated. The greatest mean seedling loss after three years of investigations when compared to the control was 76.8% (Akt), and the lowest 18.4% (Bajka). After three years of studies the loss of plants before harvest ranged from 29.0% (Bajka) to 85.7% (Akt). The mean decrease of panicle numbers compared to the control ranged from 14.7% (Bajka) to 79.4% (Akt). The 3-year means of grain yield loss, as compared to the control, were the lowest in cv. Borowiak (35.2%) and Bajka (40.1%), while the highest in cv. Akt (84.2%). On average after three years of experiments, isolates of *B. sorokiniana* collected from plants of the studied oat genotypes developed from the artificially inoculated grain accounted for 76.2% of the total number of fungi isolated from disinfected kernels and 65.9% from un-disinfected kernels. The fungi like: *A. alternata, B. sorokiniana, F. culmorum, F. graminearum, F. oxysporum, F. poae, F. sporotrichioides* and *P. verrucosum* var. *cyclopium* from both disinfected and un-disinfected control kernels were isolated the most frequently.

Key words: *Bipolaris sorokiniana*, genotypes, oat, pathogenicity

INTRODUCTION

So far, Polish studies of the occurrence of fungal diseases in oat (*Avena sativa* L.), show that this cereal is commonly infected by pathogens of the genus *Fusarium* (Kiecana 1998, Kiecana, Mielniczuk 2001, Kiecana *et al.* 2002, 2003, 2005, Mielniczuk *et al.* 2004). In contrast, there is little information on the pathogenicity of *Bipolaris sorokiniana* in relation to oats (Scott 1995, Clear *et al.* 2000 b).

Bipolaris sorokiniana (Sacc.) Shoem. plays an important role in infections of various cereals (Łacicowa *et al.* 1993, Clear *et al.* 2000 a, b, Wiśniewska *et al.* 1998). The occurrence of *B. sorokiniana* in naked oat grain in central and southern parts of the Lublin province was reported by Łacicowa (1967), while Mielniczuk (1999) described the occurrence of this pathogen in oat grain and

Communicated by Edward Arseniuk

hulls from the north-eastern parts of the province. This fungal species was isolated also from roots and leaf sheaths of oat plants cultivated in that region (Kiecana *et al.* 2003). In the 1950s, Cuturilo (1951, according to Arsenijević *et al.* 1996) reported that *B. sorokiniana* infected oats in Yugoslavia.

Bipolaris sorokiniana can cause seedling blight in cereals, including oats in the first weeks of growth (Arabi, Jawhar 2004, Liljeroth *et al.* 1996). In older plants this pathogen can cause crown rot, spots on leaves, and pathogenic changes in the colour of hulls. The fungus delays plant development, which affects total and productive tillering, lowers grain yield, and causes a darker pigmentation of embryos in oat grain (Łacicowa, Pięta 1991, Valjavec-Gratian, Steffenson 1997, Duveiller, Garcia Altamirano 2000, Hossain, Hossain 2001, Kiecana *et al.* 2003, Fernandez, Jefferson 2004).

The pathogenicity of *B. sorokiniana* is associated with the production of secondary metabolites, namely sorokinianin and prehelminthosporol, which are phytotoxic (Nakajima *et al.* 1998). This species produces also other toxic secondary metabolites: victoxinin and helminthosporol (Kachlicki 1995). The correlation between pathogenicity of strains of *B. sorokiniana* and their ability to produce phytotoxins was noted by Ludwig (1957).

Moreover, in the case of *B. sorokiniana*, an ability to produce anthraquinone compounds (helminthosporin and cynodontin) was observed. They play the role of autoregulators of growth as well as bacterial inhibitors (Tsurushima *et al.* 1984, according to Engström *et al.* 1993).

According to Rabie *et al.* (1976) *B. sorokiniana* is able to produce sterigmatocystin, which is carcinogenic to warm-blooded organisms.

The high harmfulness of *B. sorokiniana* to barley and wheat (Łacicowa, Pięta 1998, Łacicowa *et al.* 1993, Liljeroth *et al.* 1996, Valjavec-Gratian, Steffenson 1997, Almgren *et al.* 1999, Clear *et al.* 2000 a) and the proved unequal sensitivity of various cereal species to root and crown infection by this pathogen (Łacicowa, Pięta 1991, Łacicowa *et al.* 1993), suggested the possibility of similar variation also in the case of oat genotypes and inclined us to study this variation.

MATERIAL AND METHODS

The investigations were carried out in 2000-2002 on experimental plots in Zamość region (south-eastern part of Poland), on a leached brown soil, formed on loess deposits (FAO 1998), where root crops had been grown previously. Every year the recommended rates of NPK fertilization (Mazurek *et al.* 1993) and manual weeding were applied.

In this study *B. sorokiniana* isolate No. 36 from oat grain, which in laboratory tests according to the method of Mishra and Behr (1976) showed the highest pathogenicity was used. Grain samples of 12 oat genotypes were obtained from Plant Breeding Companies in Choryń and Strzelce (Table 1).

In each year of this study, the experiment included a block of plots sown with grain artificially inoculated with *B. sorokiniana*, and a block of control plots sown with uninoculated grain. The methods of inoculation, mixture preparation and grain inoculation before sowing, as well as experimental design and methods of observations, were the same as in paper on triticale (Łacicowa, Kiecana 1986).

The results were analysed statistically by using the Tukey test and confidence half-intervals (Żuk 1989).

The mycological analysis of grain collected from plants developed from artificially inoculated grain and from control grain was made on a maltose medium (Extrait de malt bioMérieux) with added streptomycin (1 mg × l^{-1}). For each genotype, in each experimental combination, 100 kernels (50 un-disinfected and 50 disinfected for 1 min in 50% C_2H_5OH and for 1 min in 0.1% $HgCl_2$) were analysed. The kernels in Petri dishes were incubated in a termostate for 8 days at 22°C. The obtained colonies of fungi after obtaining to one-spore cultures were identified to the species on PDA medium (Difco) after 14 days of keeping the cultures at the temperature 24şC with no light access. The cultures of fungi were identified to the species using monographs and keys by Booth (1971), Domsch *et al.* (1980), Ellis (1971), Gilman (1957), Munk (1957), Nelson *et al.* (1983), Ramirez (1982), Rifai (1969), and Thom and Raper (1945).

Information on weather conditions during this study was obtained from the Institute of Agricultural Sciences in Zamość.

RESULTS

During the 3-year field experiments, a lower number of plants developed in each growing season from grain inoculated with *B. sorokiniana* No. 36 than from the control grain. The highest loss of plants, due to pre and postemergence damping-off, was recorded 7 weeks after the artificially inoculated grain was sown. Over the 3 years, the mean percentage of seedling loss due to the inoculation, as compared to the control, was the highest in cv. Akt (76.8%) and the lowest in cv. Bajka (18.4%). (Fig. 1).

The statistical analysis of seedling numbers showed that every year the inoculation with *B. sorokiniana* caused significant losses in cultivars: Akt, Bohun, Jawor, Polar and breeding line CHD 2099 (Table 1).

Plant numbers in particular genotypes, determined on experimental plots before oat harvest in 2000, only slightly differed from the numbers recorded during the first observations, or were exactly the same. In 2001 and 2002, numbers of plants in all cultivars and breeding lines before harvest were lower than during the first observations in spring. Plant numbers before harvest in 2002 differed significantly from the control in 11 genotypes (except cv. Borowiak), whereas in 2001 in 7 oat genotypes (Table 2). After 3 years of experiments, the plant losses before harvest due to the inoculation reached on average from 29.0% (Bajka) to 85.7% (Akt) (Fig. 1).

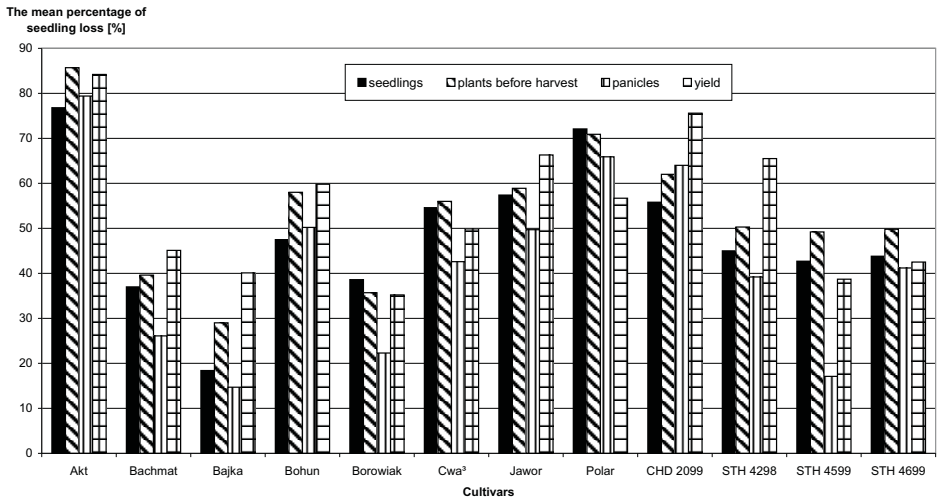

Fig. 1. The mean percentage of seedling loss, plants before harvest, panicles and yield loss due to the artificial inoculation of oat kernels by *Bipolaris sorokiniana* No. 36 after 3 years of studies 2000-2002

Table 1

The influence of artificial oat grain infection by Bipolaris sorokiniana No. 36 on seedling numbers in years 2000 – 2002

Cultivars and lines	Experimental combination							
	Bipolaris sorokiniana No. 36					Control		
	2000	2001	2002	χ^2	χ^2	2000	2001	2002
Akt	0.50*	4.75*	3.75*	3.00	12.92	6.50	21.75	10.50
Bachmat (CHD 1598)	8.25	12.00*	12.50	10.92	17.33	12.25	20.25	19.50
Bajka	7.75	13.00*	17.00	12.58	15.42	8.00	20.50	17.75
Bohun (CHD 1999)	5.25*	12.25*	11.75*	9.75	18.58	12.25	24.25	19.25
Borowiak	5.75	13.00*	12.25*	10.33	16.83	8.00	23.75	18.75
Cwał (CHD 1698)	6.50	9.00*	9.25*	8.25	18.17	8.5	24.00	22.00
Jawor	2.75*	9.50*	11.50*	7.92*	18.58	13.75	21.00	21.00
Polar (STH 4999)	0.75*	6.25*	3.75*	3.58	12.83	6.00	20.50	12.00
CHD 2099	3.75*	10.25*	11.50*	8.50*	19.25	15.00	23.25	19.50
STH 4298	4.75*	11.25	12.75*	9.58*	17.42	14.75	19.75	17.75
STH 4599	6.25	9.50*	5.00*	6.92	12.08	8.00	16.75	11.50
STH 4699	7.50	10.25*	15.00*	10.92	19.42	13.00	22.50	22.75

* - means differ significantly compared to the control when $p \leq 0,05$

The highest losses of panicles, as compared to the control, after 3 years of experiments, were recorded in cv. Akt (79.4%). Considerable losses of panicles, reaching up to 50%, were observed also in: Bohun, Polar and CHD 2099 (Fig. 1). The lowest percentage of panicle losses, as compared to the control, was found

in cv. Bajka (14.7%) and breeding line STH 4599 (17.1%) (Fig. 1). On the basis of 3-year means for the grain artificially inoculated with *B. sorokiniana*, significant differences in panicle numbers, as compared to the control, were found in 3 genotypes: Akt, CHD 2099 and STH 4699 (Table 3).

Table 2

The influence of artificial oat grain infection by *Bipolaris sorokiniana* No. 36 on plant numbers before harvest in years 2000 – 2002

Cultivars and lines	Experimental combination							
	Bipolaris sorokiniana No. 36					Control		
	2000	2001	2002	χ^2	χ^2	2000	2001	2002
Akt	0.50	1.50*	2.75*	1.58	11.08	6.50	16.25	10.50
Bachmat (CHD 1598)	7.75	7.25	8.25*	7.75	12.83	11.75	12.25	14.50
Bajka	6.00	8.25	12.75*	9.00	12.67	8.00	13.50	16.50
Bohun (CHD 1999)	5.25*	8.00*	7.25*	6.83*	16.25	12.00	20.00	16.75
Borowiak	5.75	10.50*	8.50	8.25	12.83	6.5	16.50	15.50
Cwał (CHD 1698)	5.75	6.75*	5.75*	6.08	13.83	7.75	15.75	18.00
Jawor	2.50*	7.00	8.00*	5.83*	14.17	13.00	12.50	17.00
Polar (STH 4999)	0.75*	5.25*	3.25*	3.08	10.58	5.75	15.25	10.75
CHD 2099	4.00*	5.50*	7.25*	5.58*	14.67	15.00	15.00	14.00
STH 4298	4.75*	6.75*	10.25*	7.25*	14.58	13.25	16.00	14.50
STH 4599	6.25	6.00	2.25*	4.83	9.50	7.00	10.25	11.25
STH 4699	5.75*	7.00	11.25*	8.00	15.92	12.75	15.00	20.00

*- means differ significantly compared to the control when p≤ 0,05

Table 3

The influence of artificial oat grain infection by Bipolaris sorokiniana No. 36 on panicle numbers in years 2000-2002

Cultivars and lines	Experimental combination							
	Bipolaris sorokiniana No. 36					Control		
	2000	2001	2002	χ^2	χ^2	2000	2001	2002
Akt	7.25*	5.00*	12.00*	8.08*	39.17	32.00	52.50	33.00
Bachmat (CHD 1598)	50.00	39.75	35.00	41.58	56.25	71.75	45.00	52.00
Bajka	49.25	49.00	55.00*	51.08	59.75	53.25	52.50	73.50
Bohun (CHD 1999)	39.00*	66.25*	33.00	46.08	92.50	97.50	117.75	62.25
Borowiak	29.75	70.00	30.75	43.50	56.00	34.25	76.50	57.25
Cwał (CHD 1698)	43.50	41.00	18.50*	34.33	59.83	56.00	47.75	75.75
Jawor	15.75*	40.25	42.75	32.92	65.42	83.75	44.25	68.25
Polar (STH 4999)	9.75	23.25*	14.00	15.67	46.00	31.00	79.75	27.25
CHD 2099	32.00*	34.75*	27.75*	31.50*	87.50	125.25	70.25	67.00
STH 4298	44.50*	48.75*	55.50	49.58	81.58	108.50	76.00	60.25
STH 4599	40.75	40.00	14.00	31.58	38.08	42.00	45.75	26.50
STH 4699	60.50	40.00	51.00	50.50*	85.92	100.00	69.75	88.00

* - means differ significantly compared to the control when p≤ 0,05

The grain yield in 2002 in all the analysed oat genotypes differed significantly from the control. In other years of experiments, grain yield differed significantly, as compared to the control, in 6 genotypes in 2000, whereas in 2001 only in cv. Akt (Table 4). The 3-year means of grain yield loss, as compared to the control, were the lowest in cv. Borowiak (35.2%) and Bajka (40.1%), while the highest in cv. Akt (84.2%) (Fig. 1).

Table 4

The influence of artificial oat grain infection by Bipolaris sorokiniana No. 36 on the grain yield [g] from the plot in years 2000 – 2002

Cultivars and lines	Experimental combination							
	Bipolaris sorokiniana No. 36					Control		
	2000	2001	2002	χ^2	χ^2	2000	2001	2002
Akt	10.19	2.35*	2.92*	5.15*	32.60	32.34	25.00	40.45
Bachmat (CHD 1598)	51.57*	29.70	11.69*	30.99	56.49	92.78	35.56	41.18
Bajka	65.64	28.43	26.64*	40.24	67.18	94.46	46.30	60.79
Bohun (CHD 1999)	55.33*	59.52	9.37*	41.41	102.95	171.31	87.70	49.83
Borowiak	46.82	52.70	12.28*	37.27	57.54	62.79	57.63	52.19
Cwał (CHD 1698)	57.57	25.75	7.77*	30.36	60.58	79.83	36.96	64.95
Jawor	25.52*	24.90	13.40*	21.27	63.15	111.96	25.27	52.23
Polar (STH 4999)	12.54*	40.85	3.89*	19.09	44.04	53.30	58.02	20.81
CHD 2099	45.29*	27.40	10.74*	27.81	114.09	242.71	52.18	47.39
STH 4298	57.08*	29.43	14.89*	33.80	97.98	192.15	43.83	57.96
STH 4599	66.94	22.42	9.38*	32.91	53.67	72.16	36.21	52.63
STH 4699	133.08	37.52	34.29*	68.30	118.83	193.47	56.97	106.04

* - means differ significantly compared to the control when $p \leq 0,05$

In 2000-2002, as a result of the mycological analysis of grain collected from plants of the studied oat genotypes, developed from the artificially inoculated grain, 1123 fungal colonies were isolated from disinfected kernels and 1471 from un-disinfected kernels (Table 5). On average, after 3 years of experiments, isolates of *B. sorokiniana* accounted for 76.2% of the total number of fungi isolated from disinfected kernels and 65.9% from un-disinfected kernels (Fig.2). Fungi of the genus *Fusarium*, isolated from both disinfected and un-disinfected kernels, were represented by: *Fusarium culmorum, F. equiseti, F. oxysporum, F. poae,* and *F. sporotrichioides*. In contrast, *F. avenaceum* was isolated only from disinfected kernels. Isolates of other fungi, from both disinfected and un-disinfected kernels, belonged to: *Alternaria alternata, Aspergillus flavus, A. niger, Drechslera avenae, Epicoccum nigrum, Penicillium verrucosum* var. c*yclopium, Sordaria fimicola, Trichoderma polysporum, T. viride* and non-sporulating forms. Moreover, from disinfected kernels: *Chaetomium elatum, Mucor hiemalis, Trichothecium roseum*; while from un-disinfected kernels: *Cladosporium cladosporioides* and *Talaromyces flavus* were isolated (Table 5).

Table 5

Fungi isolated from disinfected and un-disinfected kernels of the studied oat genotypes developed from the artificialy inoculated grain with *B. sorokiniana* No. 36 in years 2000 – 2002

Fungi species	Number of isolates						Together		Total number of isolates	
	2000		2001		2002					
	a	b	a	b	a	b	a	b	a	b
1	2	3	4	5	6	7	8	9	10	11
Alternaria alternata (Fr.) Keissler	(0) 0	(6) 2	(9) 8	(10) 8	(3) 4	(0) 0	(12) 12	(16) 10	26	26
Aspergillus flavus Link	(0) 0	(2) 4	(0) 1	(0) 0	(0) 0	(0) 0	(0) 1	(2) 4	1	6
Aspergillus niger van Tieghem	(3) 2	(0) 1	(0) 0	(3) 0	(1) 1	(3) 6	(4) 3	(6) 7	7	13
*Bipolaris sorokiniana (*Sacc.) Shoem.	(146) 187	(120) 159	(100) 125	(138) 205	(123) 175	(140) 207	(369) 487	(398) 571	856	969
Chaetomium elatum Kunze & Schmidt	(0) 0	(0) 0	(0) 1	(0) 0	(0) 0	(0) 0	(0) 1	(0) 0	1	0
Cladosporium cladosporioides (Fresen.) de Vries	(0) 0	(1) 0	(0) 0	(0) 0	(0) 0	(0) 0	(0) 0	(1) 0	0	1
Drechslera avenae (Eidam) Scharif	(1) 1	(19) 3	(7) 6	(13) 8	(1) 1	(2) 0	(9) 8	(34) 11	17	45
Epicoccum nigrum Link ex Link	(6) 5	(12) 7	(0) 0	(13) 4	(2) 3	(14) 14	(8) 8	(39) 25	16	64
Fusarium avenaceum (Fr.) Sacc.	(1) 0	(0) 0	(0) 0	(0) 0	(0) 0	(0) 0	(1) 0	(0) 0	1	0
*Fusarium culmorum (*W. G. Sm.) Sacc.	(2) 5	(0) 0	(0) 0	(8) 10	(1) 2	(0) 0	(3) 7	(8) 10	10	18
Fusarium equiseti (Corda) Sacc.	(0) 0	(2) 2	(1) 2	(0) 0	(0) 0	(12) 1	(1) 2	(14) 3	3	17
Fusarium oxysporum Schlecht.	(5) 4	(0) 0	(0) 0	(5) 9	(0) 4	(0) 0	(5) 8	(5) 9	13	14
Fusarium poae (Peck.) Wollenw.	(0) 0	(0) 2	(1) 3	(0) 0	(0) 0	(0) 0	(1) 3	(0) 2	4	2
Fusarium sporotrichioides Sherb.	(11) 6	(2) 0	(1) 0	(5) 4	(7) 5	(5) 0	(19) 11	(12) 4	30	16
Mucor hiemalis Wehmer	(0) 0	(0) 0	(1) 1	(0) 0	(0) 0	(0) 0	(1) 1	(0) 0	2	0
Papularia sphaerosperma (Persoon) von Höhnel	(0) 0	(0) 0	(2) 0	(0) 0	(0) 0	(0) 0	(2) 0	(0) 0	2	0

Numbers in brackets indicate fungi isolates obtained from germinating kernels, numbers outside the brackets fungi isolates obtained from non-germinating kernels
a-disinfected kernels, b-un-disinfected kernels

Table 5

Fungi isolated from disinfected and un-disinfected kernels of the studied oat genotypes developed from the artificialy inoculated grain with *B. sorokiniana* No. 36 in years 2000 – 2002 (continued)

Fungi species	Number of isolates						Together		Total number of isolates	
	2000		2001		2002					
	a	b	a	b	a	b	a	b	a	b
1	2	3	4	5	6	7	8	9	10	11
Penicillium verrucosum Dierckx var. *cyclopium* (West.) Samson, Stolk et Hadlok	(13) 10	(2) 9	(1) 0	(22) 12	(18) 15	(4) 8	(32) 25	(28) 29	57	57
Sordaria fimicola (Rob.) Ces. De & Not.	(0) 0	(8) 22	(8) 16	(0) 0	(0) 0	(28) 11	(8) 16	(36) 33	24	69
Talaromyces flavus Klöcker, Stolk & Samson	(0) 0	(0) 0	(0) 0	(0) 0	(0) 0	(0) 2	(0) 0	(0) 2	0	2
Trichoderma polysporum (Link ex Pers.) Rifai	(0) 0	(6) 9	(8) 2	(0) 0	(0) 0	(0) 0	(8) 2	(6) 9	10	15
Trichoderma viride Rifai	(8) 4	(5) 8	(0) 0	(15) 22	(11) 3	(13) 33	(19) 7	(33) 63	26	96
Trichothecium roseum Link	(0) 0	(0) 0	(2) 0	(0) 0	(0) 0	(0) 0	(2) 0	(0) 0	2	0
Forms not producing spores	(4) 3	(4) 4	(4) 3	(8) 7	(2) 1	(11) 7	(10) 7	(23) 18	17	41
Together	(200) 227	(189) 232	(145) 168	(240) 289	(169) 214	(232) 289	(514) 609	(661) 810	1123	1471

umbers in brackets indicate fungi isolates obtained from germinating kernels, numbers outside the brackets fungi isolates obtained from non-germinating kernels
a-disinfected kernels, b-un-disinfected kernels

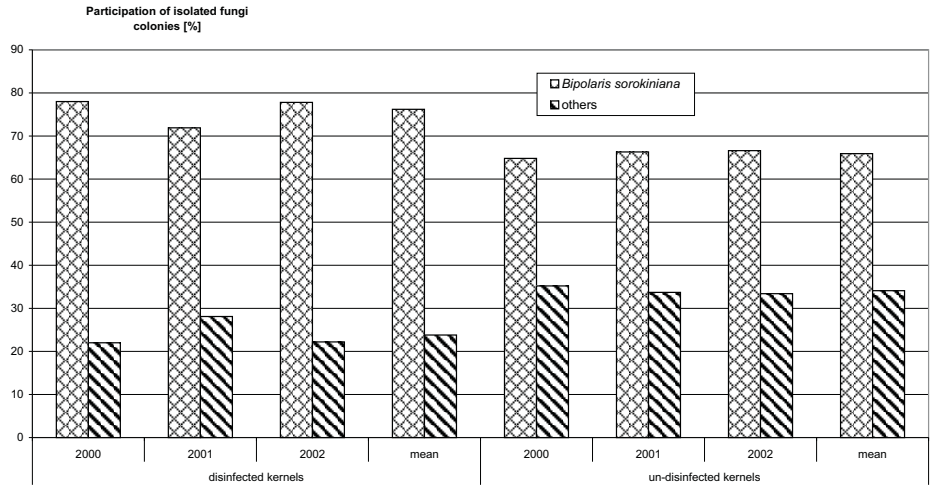

Fig. 2 The percentage participation of fungi colonies isolated from disinfected and un-disinfected kernels of the studied oat genotypes artificially inoculated by *Bipolaris sorokiniana* No. 36 in years 2000-2002

As a result of the mycological analysis of kernels collected from control plants of the studied oat cultivars and breeding lines during the 3 years, 928 fungal colonies were isolated from disinfected kernels and 1362 from un-disinfected kernels. From both disinfected and un-disinfected control kernels: *A. alternata, B. sorokiniana, F. culmorum, F. graminearum, F. oxysporum, F. poae, F. sporotrichioides* and *P. verrucosum* var. *cyclopium* were isolated the most frequently (Table 6).

In Zamość the growing season in 2000 was characterized by higher mean monthly temperatures in April, May, Jun, July and August, as compared to long-term means for those months (by 0.4-4.3°C). Monthly rainfall, compared with long-term monthly means, was higher in April, May and June (by 14-40%), but much lower in July and August. During the growing season in 2001, monthly mean air temperatures were generally higher than long-term monthly means (by 0.7-3.7°C), except in June, which was 0.4şC colder than average. In 2001 only in April and July the monthly rainfall exceeded long-term monthly means, by 27.9% and 56.8%, respectively. The growing season in 2002 was the warmest of all studied seasons, except for April. Monthly mean temperatures during the growing season in 2002 were generally higher than long-term means (by 1.9-4.6°C). Monthly rainfall exceeded long-term monthly means in May, in June and July, by: 41.8%, 69.1% and 2.5%, respectively. The lowest precipitation was recorded in August of that year (Table 7).

Table 6

Fungi isolated from disinfected and un-disinfected kernels of the studied oat genotypes developed from grain of control plants in years 2000 – 2002

Fungi species	Number of isolates						Together		Total number of isolates	
	2000		2001		2002					
	a	b	a	b	a	b	a	b	a	b
1	2	3	4	5	6	7	8	9	10	11
Alternaria alternata (Fr.) Keissler	(42) 25	(67) 43	(52) 41	(76) 44	(44) 9	(95) 45	(138) 75	(238) 132	213	370
Aspergillus flavus Link	(5) 1	(9) 3	(0) 0	(0) 0	(3) 0	(0) 0	(8) 1	(9) 3	9	12
Aspergillus niger van Tieghem	(0) 0	(0) 0	(3) 4	(4) 4	(0) 0	(0) 0	(3) 4	(4) 4	7	8
Aureobasidium pullulans de Barry Arnaud	(2) 3	(4) 3	(5) 4	(0) 0	(0) 0	(0) 0	(7) 7	(4) 3	14	7
Bipolaris sorokiniana (Sacc.) Shoem.	(24) 18	(28) 24	(25) 21	(28) 21	(24) 14	(84) 32	(73) 53	(140) 77	126	217
Cladosporium cladosporioides (Fresen.) de Vries	(0) 0	(1) 2	(0) 0	(0) 0	(0) 0	(0) 0	(0) 0	(1) 2	0	3
Drechslera avenae (Eidam) Scharif	(3) 3	(5) 4	(3) 3	(3) 2	(2) 2	(1) 0	(8) 8	(9) 6	16	15
Epicoccum nigrum Link ex Link	(5) 7	(9) 9	(9) 10	(11) 11	(1) 0	(3) 3	(15) 17	(23) 23	32	46
Fusarium avenaceum (Fr.) Sacc.	(0) 0	(2) 7	(3) 4	(4) 3	(0) 0	(0) 0	(3) 4	(6) 10	7	16
Fusarium crookwellense Burges, Nelson, Toussoun	(4) 3	(5) 5	(0) 0	(0) 0	(3) 1	(25) 4	(7) 4	(30) 9	11	39
Fusarium culmorum (W. G. Sm.) Sacc.	(14) 8	(16) 20	(11) 19	(9) 16	(7) 2	(37) 36	(32) 29	(62) 72	61	134
Fusarium equiseti (Corda) Sacc.	(1) 1	(0) 0	(2) 1	(6) 1	(0) 1	(30) 15	(3) 3	(36) 16	6	52
Fusarium graminearum Schwabe	(27) 1 3	(21) 11	(9) 5	(6) 6	(27) 8	(44) 8	(63) 26	(71) 25	89	96
Fusarium oxysporum Schlecht.	(11) 5	(17) 10	(11) 9	(13) 14	(8) 2	(6) 4	(30) 16	(36) 28	46	64
Fusarium poae (Peck.) Wollenw.	(13) 5	(11) 6	(10) 6	(7) 3	(11) 3	(11) 3	(34) 14	(29) 12	48	41

Numbers in brackets indicate fungi isolates obtained from germinating kernels, numbers outside the brackets fungi isolates obtained from non-germinating kernels
a-disinfected kernels, b-un-disinfected kernels

Table 6

Fungi isolated from disinfected and un-disinfected kernels of the studied oat genotypes developed from grain of control plants in years 2000 – 2002 (continued)

Fungi species	Number of isolates						Together		Total number of isolates	
	2000		2001		2002					
	a	b	a	b	a	b	a	b	a	b
1	2	3	4	5	6	7	8	9	10	11
Fusarium sporotrichioides Sherb.	(5) 5	(9) 12	(8) 16	(13) 18	(1) 0	(10) 10	(14) 21	(32) 40	35	72
Mucor hiemalis Wehmer	(5) 2	(4) 4	(4) 4	(9) 2	(5) 0	(1) 0	(14) 6	(14) 6	20	20
Penicillium verrucosum Dierckx var. *cyclopium* (West.) Samson, Stolk et Hadlok	(7) 8	(10) 10	(10) 9	(14) 10	(6) 5	(1) 1	(23) 22	(25) 21	45	46
Sordaria fimicola (Rob.) Ces. De & Not.	(6) 4	(11) 4	(7) 6	(0) 0	(6) 2	(0) 0	(19) 12	(11) 4	31	15
Trichoderma aureoviride Rifai	(0) 0	(5) 3	(0) 0	(0) 0	(0) 0	(0) 0	(0) 0	(5) 3	0	8
Trichoderma polysporum (Link ex Pers.) Rifai	(0) 0	(0) 0	(6) 5	(7) 3	(0) 0	(0) 0	(6) 5	(7) 3	11	10
Trichoderma viride Rifai	(0) 0	(2) 5	(7) 7	(15) 7	(0) 0	(0) 0	(7) 7	(17) 12	14	29
Trichothecium roseum Link	(2) 0	(0) 0	(0) 0	(0) 0	(0) 1	(0) 0	(2) 1	(0) 0	3	(0) 0
Forms not producing spores	(21) 10	(14) 11	(11) 10	(9) 7	(27) 5	(0) 1	(59) 25	(23) 19	84	42
Together	(197) 121	(250) 196	(196) 184	(234) 172	(175) 55	(348) 162	(568) 360	(832) 530	928	1362

Numbers in brackets indicate fungi isolates obtained from germinating kernels, numbers outside the brackets fungi isolates obtained from non-germinating kernels
a-disinfected kernels, b-un-disinfected kernels

Table 7

Air temperature and rainfall in Zamość during the growing seasons of oat 2000 – 2002

Month	Long-term monthly means (1971 -1995)		Deviations of temperatures			Percentage of long-term monthly mean rainfally		
	Temperature [°C]	Rainfall [mm]	2000	2001	2002	2000	2001	2002
April	7.3	43	+4.3	+1.0	+1.9	140.0	127.9	59.8
May	13.1	62	+1.4	+0.7	+4.6	120.0	38.7	141.8
June	16.4	81	+0.4	-0.4	+1.9	114.0	84.9	169.1
July	17.9	91	+0.9	+3.6	+4.3	21.0	156.8	102.5
August	17.0	81	+2.5	+3.7	+3.8	1.2	62.6	21.5

DISCUSSION

The method used for comparison of sensitivity of oat genotypes to *Bipolaris sorokiniana*, proved to be efficacious in experimental conditions.

In field conditions, seed germination took place in the presence of *B. sorokiniana* introduced onto the surface of hulls in the case of hulled oats, or onto the pericarp fused with the seed coat in naked oats, and to the soil environment as an inoculation mixture. In this way, a close contact of the pathogen with plants of individual oat genotypes was ensured.

Considering the variable virulence of strains within the population of *B. sorokiniana* (Valjavec-Gratian, Steffenson 1997, Almgren *et al.* 1999, Duveiller, Garcia Altamirano 2000), in field research was used only a strain whose pathogenicity tested according to the method of Mishra and Behr (1976) was the greatest. The differences in pathogenicity of strains of *B. sorokiniana*, are considered by Almgren *et al.* (1999) and Apoga *et al.* (2002) as results of unequal ability to produce phytotoxins, especially prehelminthosporol.

This study shows that plant losses on experimental plots after inoculation with *B. sorokiniana* resulted mainly from seedling death in the first 7 weeks after the artificially inoculated grain was sown. During further growth, only a small proportion of plants died, so differences in plant numbers between these observation periods were observed. That is why, in order to compare the sensitivity of cultivars, we focused on the percentage of plant losses before harvest and on grain yield per experimental plot.

In the 3 analysed growing seasons, during which the experiments were carried out, weather conditions in 2002 proved to be the most favourable for plant infection by *B. sorokiniana*, because May in that year was characterized by humid and warm weather. Similar weather conditions in 1992 were also favourable for infection of barley seedlings by *B. sorokiniana* (Łacicowa, Pięta 1998).

Considering 50% plant losses before harvest and 48% grain yield losses, on average after 3 years of experiments, the following genotypes were the most

susceptible to infection by *B. sorokiniana*: Akt, Bohun, Cwał, Jawor, Polar, CHD 2099 and STH 4298. The lowest 3-year mean of plant losses, below 30%, and mean grain yield losses only slightly exceeding 40% as a result of inoculation with *B. sorokiniana*, were recorded in cv. Bajka.

A large number of colonies of *B. sorokiniana* was isolated from the seed material collected from the plants that developed from the grain artificially inoculated with this species. This shows that grain is an important source of infection for the plants developed from it (Łacicowa 1990).

It is known from the literature that the frequency of infection of barley grain by *B. sorokiniana* depends on the amount of the inoculation material present in the plant's environment during flowering and seed formation. Most often, grain is infected by air-borne conidia from infected leaves. High humidity is particularly favourable for barley grain infection by *B. sorokiniana* (Coutre, Sutton 1978, according to Łacicowa 1990). During this study, weather conditions were favourable for infection of oat panicles and grain by *B. sorokiniana*, because May and June in 2000 and 2002 were characterized by high rainfall and air temperature, which markedly exceeded long-term means for those months.

An increase in the importance of *B. sorokiniana* in oat grain infection in Canada was reported by Clear *et al.* (2000 b). In Poland the presence of *B. sorokiniana* on grain of this species was confirmed by Łacicowa (1967) and Mielniczuk (2001). The infection of seed material by *B. sorokiniana* may be an indicator of the threat of disease to oat plantations.

The procedure of grain disinfection only partly eliminated *B. sorokiniana.* This attests to the close contact of this fungus with grain and to colonization of its inner parts. The higher frequency of isolation of *B. sorokiniana* from non-germinating kernels indicates that this pathogen lowers the seed germinability of oats and barley (Łacicowa, Pięta 1991, Mielniczuk 1999).

The fungi accompanying *B. sorokiniana* on grain were mainly the toxin-producing *Fusarium* species, such as: *F. avenaceum, F. crookwellense, F. culmorum, F. graminearum, F. equiseti, F. poae* and *F. sporotrichioides.* Studies of interactions among the fungi colonizing cereal grain show that *B. sorokiniana* is less competitive than *Fusarium* spp. (Łacicowa 1973), so appropriate media should be used for isolation of this species.

The application of the maltose medium with added streptomycin for isolation of *B. sorokiniana* proved to be effective and enabled the isolation of numerous colonies of this fungus. Apart from the maltose medium, for isolation of *B. sorokiniana* from the infected oat grain, also the filter paper test and Reis medium (Reis *et al.* 1999) can be used.

CONCLUSIONS

Among the tested oat genotypes, none was completely resistant to seedling infection by *B. sorokiniana*.

Infection of oat grain by *B. sorokiniana* can result in increased incidence of seedling blight during oat cultivation, but the incidence of oat seedling damping-off caused by this species is modified by weather conditions.

Cultivar Bajka can be recommended for use as a standard in tests of oat sensitivity to infection by *B. sorokiniana,* because its relatively high resistance to this pathogen was observed in all years of experiments.

Cultivar Akt should not be cultivated in the Zamość province, as it is highly sensitive to seedling infection by *B. sorokiniana* in the climatic and soil conditions of this region.

REFERENCES

Almgren I., Gustafsson M., Fält A.S., Lindgren H., Liljeroth E. 1999. Interaction between root and leaf disease development in barley cultivars after inoculation with different isolates of *Bipolaris sorokiniana.* J. Phytopathol. 147: 331 – 337.

Apoga D., Akesson H., Jansson H.B., Odham G. 2002. Relationship between production of phytotoxin prehelminthosporol and virulence in isolates of the plant pathogenic fungus *Bipolaris sorokiniana.* European J. of Plant Pathol. 108: 519 – 526.

Arabi M. I., Jawhar M. 2004. Repeatibility of barley seedling reaction to spot blotch and common root rot. Cereal Research Communications 32, 2: 249 – 253.

Aresnijević M., Draganić D., Kenžević T. 1996. Verste nekadašnjeg roda „*Helminthosporium*" utvrdene u Jugoslaviji (1922 – 1995). Zaštita bilja 47, 2, 216: 93 – 119.

Booth C. 1971. The genus *Fusarium.* Comm. Mycological Institute Kew, Surrey England.

Clear R.M., Patrick S.K., Gaba D. 2000 a. Prevalence of fungi and fusariotoxins on barley seed from western Canada, 1995 to 1997. Can. J. Plant Pathol. 22: 44 – 50.

Clear R.M., Patrick S.K., Gaba D. 2000 b. Prevalence of fungi and fusariotoxins on oat seed from western Canada, 1995 – 1997. Can. J. Plant Pathol. 22: 310 – 314.

Domsch K.H., Gams W., Anderson T.H. 1980. Compendium of soil fungi. Academic Press London, New York, Toronto, Sydney, San Francisco.

Duveiller E., Garcia Altamirano I. 2000. Pathogenicity of *Bipolaris sorokiniana* isolates from wheat roots, leaves and grains in Mexico. Plant Pathol. 49: 235 – 242.

Ellis M.B. 1971. Dematiaceous Hyphomycetes. Commonwealth Mycological Institute, Kew Surrey, England.

Engström K., Brishammar S., Svensson Ch., Bengtsson M., Andersson R. 1993. Anthraquinones from some *Drechslera* species and *Bipolaris sorokiniana.* Mycol. Res. 97 (3): 381 – 384.

FAO 1998. World reference base for soil resources. Food and Agriculture Organization of the United Nations, Rome, Italy.

Fernandez M.R., Jefferson P.G. 2004. Fungal populations in roots and crowns of common and durum wheat in Saskatchewan. Can. J. Plant Pathol. 26: 325 – 334.

Gilman J.C. 1957. A manual of soil fungi. The Iowa State College Press-Ames, Iowa, USA.

Hossain M.M., Hossain I. 2001. Effect of Black Pointed Seed in Seed Sample on Leaf Spot Severity and Grain Infection of Wheat in the Field. Pakistan J. of Biological Sciences 4, 11: 1350 – 1352.

Kachlicki P. 1995. Metabolites of Helminthosporia. W: Chełkowski J. Helminthosporia metabolites, biology, plant diseases *Bipolaris*, *Drechslera*, *Exserohilum*: 1 – 26.

Kiecana I. 1998. Występowanie *Fusarium* spp. na owsie (*Avena sativa* L.). Postępy w Ochronie Roślin Poznań. 38, 2: 541 – 543.

Kiecana I., Mielniczuk E. 2001. Występowanie *Fusarium culmorum* (W.G.Sm.) Sacc., *Fusarium avenaceum* (Fr.) Sacc. oraz *Fusarium crookwellense* Burgess, Nelson & Toussoun na rodach hodowlanych owsa (*Avena sativa* L.). Acta Agrobotanica 64, 1: 83 – 93.

Kiecana I., Mielniczuk E., Kaczmarek Z., Kostecki M., Goliński P. 2002. Scab response and moniliforim accumulation in kernels of oat genotypes inoculated with *Fusarium avenaceum* in Poland. Europ. J. Plant Pathol., 108: 245 – 251.

Kiecana I.,. Mielniczuk E., Cegiełko M., Pszczółkowski P. 2003. Badania nad chorobami podsuszkowymi owsa (*Avena sativa* L.) z uwzględnieniem temperatury i opadów. Acta Agrobotanica 56, 1-2: 95 – 107.

Kiecana I., Mielniczuk E. Perkowski J., Goliński P. 2005. Porażenie wiech przez *Fusarium poae* (Peck) Wollenw. oraz zawartość mikotoksyn w ziarnie owsa. Acta Agrobotanica 58, 2: 91 – 102.

Liljeroth E., Franzon-Almgren I., Gunnarsson T. 1996. Root Colonization by *Bipolaris sorokiniana* in Different Cereals and Relations to Lesion Development and Natural Root Cortical Cell Death. J. Phytopathol. 144: 301 – 307.

Ludwig R. A. 1957. Toxin production by *Helminthosporium sativum* P. K. & B. and its significance in disease development. Can. J. Bot. 35: 291 – 303.
Łacicowa B. 1967. Badania mikoflory materiału siewnego owsa uprawianego na obszarze województwa lubelskiego. Annales UMCS, sec. E, XXII, 13: 197 – 206.
Łacicowa B. 1973. Wzajemne oddziaływanie niektórych grzybów zasiedlających materiał siewny zbóż. Acta Mycol. IX, 1: 7 – 10.
Łacicowa B. 1990. Mikoflora ziarna jęczmienia jarego (*Hordeum vulgare* L.) wzrastającego w warunkach zagrożenia chorobowego przez *Drechslera sorokiniana* (Sacc.) Subram et Jain. (=*Helminthosporium sativum* P.K. et B.). Rocz. Nauk Roln. sec. E, 20, ″: 17 – 23.
Łacicowa B., Kiecana I. 1986. Występowanie *Fusarium nivale* (Fr.) Ces. na pszenżycie i podatność różnych rodów hodowlanych na porażenie. Rocz. Nauk. Rol. ser. E., 9, 1: 93 – 113.
Łacicowa B., Pięta D. 1991. Podatność różnych odmian jęczmienia jarego na porażenie przez *Drechslera sorokiniana* (Sacc.) Subram. et Jain. Hod. Rośl. Aklim. 35, 5/6: 53 – 59.
Łacicowa B., Pięta D. 1998. Wpływ temperatury i opadów na udział grzybów w powodowaniu chorób podsuszkowych jęczmienia jarego (*Hordeum vulgare* L.). Acta Agrobotanica 51, (1 – 2): 51 – 61.
Łacicowa B., Pięta D., Kiecana I., Kukiełka R. 1993. Ochronne działanie zapraw chemicznych w warunkach zagrożenia jęczmienia jarego (*Hordeum vulgare* L.) przez choroby podsuszkowe. Mat. XXXIII Sesji Nauk. IOR cz. I, Referaty: 195 – 201.
Mazurek J., Mazurek J., Król M. 1993. Wpływ odmiany gleby i agrotechniki na plonowanie owsa. W: Biologia i agrotechnika owsa. Puławy, IUNG: 247 – 308.
Mielniczuk E. 1999. Badania nad fuzariozą wiech owsa (*Avena sativa* L.) z uwzględnieniem podatności odmian i zawartości mikotoksyn w ziarnie. Praca doktorska.
Mielniczuk E. 2001. The occurrence of *Fusarium* spp. on panicles of oat (*Avena sativa* L.). J. Plant Protection Research 41, 2: 173 – 180.
Mielniczuk E., Kiecana I., Perkowski J. 2004. Susceptibility of oat genotypes to *Fusarium crookwellense* Burgess, Nelson and Toussoun infection and mycotoxin accumulation in kernels. Biologia, Bratislava 59/6: 809 – 816.
Mishra C.B.P., Behr L. 1976. Der Einfluss von *Fusarium culmorum* (W.G.Sm.) Sacc., *Fusarium avenaceum* (Fr.) Sacc. und *Fusarium nivale* (Fr.) Ces., *Griphosphaeria nivalis* Müller et Arx auf die Keimung des Weixen. Arch. Phytopahol. Pflanzenschutz 12, 6: 373 – 377.
Munk A. 1957. Danish *Pyrenomycetes*. A. Preliminary Flora. Copenhagen Ejnar Munksgaard.
Nakajima H., Toratsu Y., Fujii Y., Ichinoe M., Hamasaki T. 1998. Biosynthesis of Sorokinianin a Phytotoxin of *Bipolaris sorokiniana*: Evidence of Mixed Origin form the Sesquiterpene and TCA Pathways. Tetrahedron Letters, 39: 1013 – 1016.
Nelson P.E., Toussoun T.A., Marasas W.F.O. 1983. *Fusarium* species. An Illustrated Manual for Identification. The Pennsylvania State University Press University Park and London.
Rabie C.J., Lubben A., Steyn M. 1976. Production of sterigmatocystin by *Aspergillus versicolor* and *Bipolaris sorokiniana* on Semisynthetic Liquid and Solid Media. Applied and Environmental Microbiology 32, 2: 206 – 208.
Ramirez C. 1982. Manual and atlas of the Penicillia. Elsevier Biomedical Press., Oxford.
Reis E.M., Reis A.C., Casa R.T., Blum M.M.C. 1999. Comparison of methods to deteact leaf and head blight fungi in small grains seeds. Summa Phytopathologica 25: 364 – 367.
Rifai M.A. 1969. A revision of the genus *Trichoderma.* Kew, Surrey, England.
Scott D.B. 1995. Helminthosporia that cause leaf spots on small-grain cereals in South Africa. in: Chełkowski J. Helminthosporia metabolites, biology, plant diseases *Bipolaris, Drechslera, Exserohilum*.
Thom Ch., Raper K.B. 1945. A manual of the Aspergilli Baltimore. The Williams & Wilkins Company.
Valjavec-Gratian M., Steffenson B.J. 1997. Pathotypes of *Cochliobolus sativus* on Barley in North Dakota. Plant Dis. 81, 11: 1275 – 1278.
Wiśniewska H., Wakuliński W., Chełkowski J. 1998. Susceptibility of barley to *Bipolaris sorokiniana.* Seedling blight determined by disease sloving and electrolyte leakage. J. Phytopathol. 146: 563 – 566.
Żuk B. 1989. Biometria stosowana. PWN. Warszawa.