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FUNGI INFECTING THE ROOTS AND STEM BASE OF WINTER RYE (*Secale cereale* L.) GROWN IN THE LUBLIN REGION (POLAND)

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Abstract

This study was carried out in the period 2001-2005 in 10 winter rye plantations located in the Lublin region. The percentage of winter rye stems with necrosis symptoms on the lower internodes ranged from 32.5% to 71% in 2001, 38% to 68% in 2002, 21.5% to 56.5% in 2003, 35% to 70% in 2004, and 36% to 88% in 2005. The mean values of the disease index ranged from 7.5 to 46.75 and they differed significantly. The results of mycological analysis showed that the main pathogens infecting the roots and stem base of rye were the species *Fusarium avenaceum* (24% of all isolates) and *F. culmorum* (25% of all isolates).

Key words: *Fusarium* spp., winter rye, root rot, foot rot

INTRODUCTION

In Poland rye cultivation is extensive. Moreover, this plant has relatively low soil and water requirements as well as it is characterized by low sensitivity to forecrop and to acidic soil reaction. Therefore, it still occupies a large acreage in spite of the declining trend that has been observed for several years. Rye grain is used for many purposes. According to estimates, as much as half of annual crop production is used for animal feed purposes, despite that this type of fodder is not the best for most groups of animals. Besides, rye grain is used for flour and alcohol production. Starch is an important ingredient of rye flour and its properties determine the usefulness of this flour for bread baking (Małecka, 2005).

Population varieties enjoy greater popularity in our country and among them the cultivar 'Dańkowskie Złote' has held the dominant position for many years. On the other hand, the proportion of hybrid varieties in crop area is still small, in spite of the fact that they surpass population varieties in productivity. However, they are more susceptible to infection by pathogens, mainly from the order Uredinales, as well as to infection of seedlings and ears by *Fusarium* spp. (Małecka, 2005; Kiecana et al. 2009; Kiecana and Mielniczuk, 2010).

Rye has a stimulating effect on the development of antagonistic bacteria to *Fusarium* spp., but species of this genus are considered to be the most harmful to rye crops (Kurek and Jaroszuk-Ściseł, 2003; Głażewska-Maniewska et al. 2004, Pałys et al. 2004; Kiecana et al. 2009). The risk of development of stem base diseases of cereals, including rye, caused by the above-mentioned pathogens increases with the introduction of reduced tillage systems without deep ploughing (Kurowski et al. 2008; Piąskowska et al. 2010). In recent years, there has been observed a particularly high proportion of the species *F. culmorum* in causing foot rot of cereals (Pałys et al. 2004; Kiecana et al. 2008, 2009; Kurowski et al. 2008). The pathogenicity of this fungus is associated with the production of toxic secondary metabolites (Logrieco et al. 2003; Šrobárová et al. 2008). Furthermore, the species *F. equiseti* and *F. sporotrichioides* participate more and more frequently in infection of seedlings, roots and stem bases (Kiecana et al. 2008; Kiecana and Mielniczuk, 2010). The main sources of infection with diseases caused by *Fusarium* spp. are the soil and seed material (Logrieco et al. 2003; Mielniczuk et al. 2010).

The present research was undertaken due to the scarcity of studies on fungi damaging the roots and stem base of winter rye grown in the Lublin region.

MATERIALS AND METHODS

This study was carried out in the period 2001-2005 in 10 winter rye plantations located in the Lublin region. A list of plantations and cultivars is given in Table 1. Infection of the stem base of rye at the hard dough stage (87 in Tottman's scale, 1987) was evaluated in each growing season. To this end, 200 stems were sampled – 4 x 50 in different places of each plantation. The sampling method was the same as in a study on oats conducted by Kiecana et al. (2008).

In the laboratory, the percentage of stems with symptoms of necrotic streaks on the lower internodes was estimated and the degree of stem infection was determined using the following scale:

0° – no symptoms
1° – up to 10% of the stem base area infected
2° – from 11% to 25% of the stem base area infected
3° – from 26% to 50% of the stem base area infected
4° – from 51% to 75% of the stem base area infected
5° – from 76% to 100% of the stem base area infected.

Next, the disease index was calculated for individual replications using Mc Kinney's formula (Łacicowa, 1969) and subsequently the average disease index was determined for each plantation, similarly as in the above-mentioned study on oats (Kiecana et al. 2008). The obtained results were statistically analysed using Tukey's confidence half-intervals (Żuk, 1989).

Mycological analysis of diseased stems and roots was performed in the laboratory. In each year of the study, 50 root fragments and 50 fragments from the lower internodes of infected stems were analysed for each plantation. Mineral medium was used to isolate fungi (Mielniczuk et al. 2010). The fungal colonies obtained were identified to the level of species using the studies such as that by Kiecana et al. (2011).

RESULTS

In each growing season, there were rye stems with symptoms of necrotic streaks on the lower internodes. The percentage of such stems ranged from 32.5% to 71% in 2001, 38% to 68% in 2002, 21.5% to 56.5% in 2003, 35% to 70% in 2004, and 36% to 88% in 2005 (Fig. 1). The mean disease index determined for rye stems sampled from the plantations in question ranged from 7.5 to 46.75 and they differed significantly between years (Table 1).

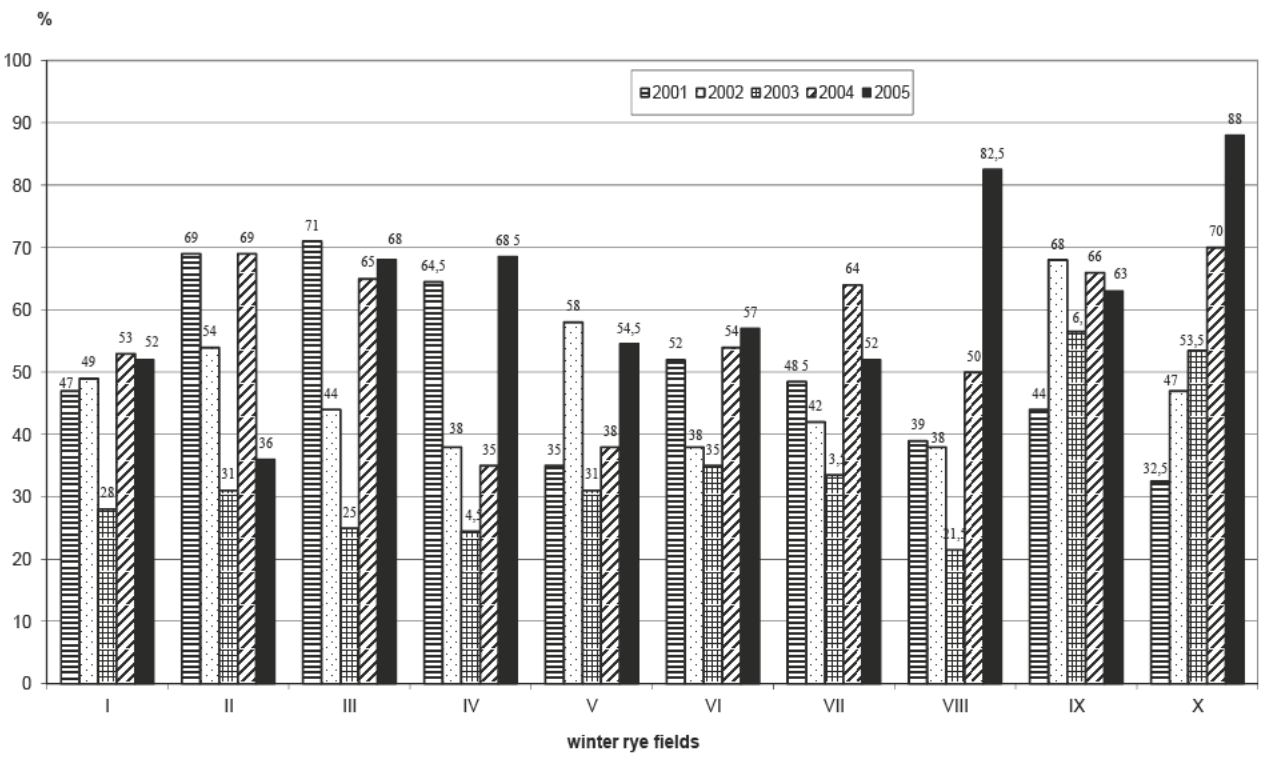

Fig. 1. Percentage of rye stem bases with disease symptoms.

Table 1
Values of the disease index for roots and stem bases of rye grown in the Lublin region in 2001-2005

Plantation number	Year				
	2001	2002	2003	2004	2005
I	21.40ab	19.70ab	11.75bc	28.25d	25.75b
II	34.90d	22.62b	13.88c	30.20d	16.50a
III	36.10d	17.65ab	8.25a	30.13d	39.50c
IV	28.70c	16.25a	7.5a	15.55a	41.50c
V	22.10b	21.95b	10.75b	23.60c	18.00a
VI	26.10b	15.70a	16.5d	19.10b	26.00b
VII	22.30b	24.00bc	14.50c	33.45e	18.75a
VIII	19.00ab	16.60a	7.75a	22.10c	44.25d
IX	16.40a	27.95c	25.50f	37.20f	24.50b
X	15.55a	20.00ab	19.75e	37.45f	46.75d
Mean	24.26	20.24	13.55	27.70	30.75
NIR/LSD	5.96	4.6	1.74	2.56	2.61

The winter rye fields analyzed were located in:
I, II, III – Wólka Zabłocka; IV,V – Brzostówka; VI,VII,VIII – Kol. Wola Sernicka; IX – Głębokie, X – Krasne
Mean values in columns followed by the same letter do not differ significantly at $P \leqslant 0.05$

As a result of mycological analysis of diseased stems and roots, 3964 fungal isolates, including 1732 isolates from the roots and 2232 from the lower internodes, were isolated over the five-year study period (Table 2). Among the isolated fungal colonies, *Fusarium* spp. accounted for 68.3%; isolates of these fungi constituted 64.7% of isolates from the roots and 71.1% of fungal colonies obtained from the lower internodes. The species *F. avenaceum* was obtained in largest numbers from the roots – 23.6% of all fungi isolated from the roots, while *F. culmorum* colonies were predominant among those obtained from the stem base – 27.2% of all isolates from the lower internodes of the stems. Irrespective of the growing season, the species *F. avenaceum* and *F. culmorum* were isolated in greatest numbers both from the roots and lower internodes; in 2003 the species *F. avenaceum* was represented in greatest numbers, since it accounted for 26.6% of all isolates, whereas *F. culmorum* colonies were 6.2% of all fungal isolates in this study year. In 2005 the percentage of *F. avenaceum* and *F. culmorum* isolates was 17.7% and 33.7%, respectively, of all fungal colonies obtained. In the other study years, the proportions of the species *F. avenaceum* and *F. culmorum* in the total number of isolates obtained both from the roots and the stem base were similar (Table 2). The species *F. equiseti* and *F. oxysporum* were also isolated from the infected plants in all growing seasons; their isolates accounted for 5.0% and 6.9%, respectively, of all isolates over the five-year study period. The genus *Fusarium* was represented by *F. graminearum, F. poae*, and *F. sporotrichioides* (Table 2).

In the years 2002, 2004 and 2005, *Bipolaris sorokiniana* was isolated both from the roots and the stem base – 67 isolates (1.7% of all isolates after the five-year study period), while in the years 2001, 2002, 2004 and 2005 the species *Rhizoctonia solani* was isolated – 73 isolates (1.8% of all fungi) (Table 2). Colonies of other fungi belonged to the following: *Alternaria alternata* (3.1% of all fungal colonies isolated over the five-year study period), *Aureobasidium pullulans* (13.9%), *Epicoccum nigrum* (1.0%), *Penicillium verrrucosum* var. *cyclopium* (0.7%), *Stemphylium botryosum* (0.8%), *Trichoderma aureoviride* (3.3%), *Trichoderma viride* (1.5%), *Talaromyces flavus* (0.6%), and non-sporulating forms (2.9%) (Table 2).

Table 2
Fungi isolated from rye roots and stem bases in 2001-2005

Fungus species	Number of isolates in each study year										Total isolates (%)		Total number of isolates (%)
	2001		2002		2003		2004		2005				
	r	sb	r	sb	r	sb	r	sb	r	sb	r	sb	
Alternaria alternata (Fr.) Keissler	32	14	11	12	6	7	15	11	4	12	68 (3.9)	56(2.5)	124 (3.1)
Aureobasidium pullulans (de Bary) Arnaud	67	74	46	78	51	61	61	45	23	45	248(14.3)	303(13.6)	551 (13.9)
Epicoccum nigrum Link.	1	4	11	0	3	4	-	-	7	11	22(1.3)	19(0.8)	41 (1.0)
Bipolaris sorokiniana (Sacc.) Shoem.	-	-	9	11	-	-	23	11	2	11	34(1.9)	33(1.5)	67 (1.7)
Fusarium avenaceum (Fr.) Sacc.	53	96	45	125	72	152	146	118	92	62	408(23.6)	553(24.8)	961(24.2)
Fusarium culmorum (W.G.Sm.) Sacc.	49	85	58	126	5	47	115	212	156	137	383(22.1)	607(27.2)	990 (25.0)
Fusarium equiseti (Corda) Sacc.	22	11	7	4	18	57	2	7	34	35	83(4.8)	114(5.1)	197 (5.0)
Fusarium graminearum Schwabe	2	-	-	-	4	0	6	12	-	-	12(0.7)	12(0.5)	24 (0.6)
Fusarium oxysporum Schlecht.	12	8	15	13	119	49	8	12	16	22	170(9.8)	104(4.7)	274 (6.9)
Fusarium poae (Peck) Wollenw.	-	-	23	46	-	-	-	-	2	42	25(1.4)	88(3.9)	113 (2.9)
Fusarium sporotrichioides Sherb.	3	14	15	6	10	36	-	-	11	52	39(2.3)	108(4.8)	147 (3.7)
Mucor hiemalis Wehmer	1	2	-	-	3	7	-	1	3	0	7(0.4)	10(0.4)	17 (0.4)
Penicillium verrucosum Dierckx var. *cyclopium* (Westling.) Samson et al.	3	0			1	-	7	12	1	3	12(0.7)	15(0.7)	27 (0.7)
Rhizoctonia solani Kühn	-	1	17	4	-	-	8	24	11	8	36(2.1)	37(1.7)	73 (1.8)
Stemphylium botryosum Wallroth	13	3	1	13			-	-	-	-	14(0.8)	16(0.7)	30 (0.8)
Trichoderma aureoviride Rifai	13	17	-	-	51	11	-	17	17	3	81(4.7)	48(2.2)	129 (3.3)
Trichoderma viride Rifai			12	4			5	32	8	-	25(1.4)	36(1.6)	61 (1.5)
Talaromyces flavus (Kőcker) Stolk et Samson	1	4	-	-			-	12	4	2	5(0.3)	18(0.8)	23 (0.6)
Non- sporulating forms	6	9	7	9	19	22	7	2	21	13	60(3.5)	55(2.5)	115 (2.9)
Total	**278**	**342**	**277**	**451**	**362**	**453**	**403**	**528**	**412**	**458**	**1732**	**2232**	**3964 (100)**

r – roots; sb – stem base

DISCUSSION

The results of the present study show a high proportion of stems with disease symptoms in the winter rye plantations located in the northern part of the Lublin region. The values of the disease index recorded for rye stems were higher than those for oats (Kiecana et al. 2008), for common and durum wheat grown under soil and climatic conditions of the Lublin region (Kiecana et al. 2011), for oats grown in the Wielkopolska (Greater Poland) region (Kiecana et al. 2008) as well as for triticale grown in Dolny Śląsk Lower Silesia (Pląskowska et al. 2010).

Infection symptoms in the form of necrotic streaks were observed mostly on the lower internodes of rye stems, similarly as in the case of oats, winter wheat and spring barley grown in different areas of the Lublin region (Łacicowa et al. 1997; Pałys et al. 2004; Kiecana et al. 2008). Species of the genus *Fusarium*, in particular *F. culmorum* and *F. avenaceum*, proved to be the pathogenic factors causing this type of disease symptoms and they are also recognised to be dangerous rye pathogens at earlier growth stages (Kiecana et al. 2009). These species are also considered to be the main cause of foot rot and root rot of various cereal species grown across the country (Łacicowa et al. 1997; Pałys et al. 2004; Kiecana et al. 2008; Kurowski et al. 2008; Majchrzak et al. 2008; Pląskowska et al. 2010).

Fusarium avenacem has proved to be a more dangerous pathogen of the stem base of triticale grown in Lower Silesia (Pląskowska et al. 2010) and of rye seedlings grown in the Lublin region than *F. culmorum* (Kiecana et al. 2009). Post-harvest residues are considered to be a serious source of infection with cereal diseases caused by this species (Fernandez et al. 2009). This fungus has also been found to be characterized by significant pathogenicity to cereal seedlings, including rye, in phytotron studies with artificial infection of growing medium and kernels (Mańka 1998; Fernandez and Chen, 2005; Kiecana et al. 2009). Moniliformin takes part in the pathogenesis of diseases induced by *F. avenaceum* and it causes cell division disturbances in infected plant organs, in particular roots (Packa, 1997).

Fusarium culmorum causes root rot, seedling blight and stem base necrosis in many plant species; it is also a recognised causal agent of *Fusarium* head blight in cereals, including rye, grown in Europe, Canada, and China (Kiecana, 1994; Łacicowa et al. 1997; Kiecana et al. 2008; 2009; Strausbaugh et al. 2004; Šrobarová et al. 2008, Fernandez et al. 2009; Kiecana and Mielniczuk, 2010). Alongside crop plants, hosts of *F. culmorum* can also include weeds, among others such as: *Capsella bursa-pastoris, Galium aparine, Matricaria* spp., *Ranunculus caris, R. regens, Rumex obtusifolius, Urtica dioica, Viola arvensis*, which are a source of infection for cereals (Shaner, 2003). This fungus has also been isolated from the grain of many cereal plants grown in Poland, which indicates that seed material infection is a serious source of stem base diseases caused by *F. culmorum* in cereals (Narkiewicz-Jodko et al. 2008; Mielniczuk et al. 2010).

The great harmfulness of *F. culmorum* to cereal plants is associated with the production of phytotoxic metabolites, in particular deoxynivalenol and nivalenol, which affect the permeability of the cytoplasmic membranes of infected cells as well as protein biosynthesis and cell divisions (Dahleen and Mc Cormik, 2001; Logrieco et al. 2003).

A species that was frequently isolated from the roots and lower internodes of rye was also *F. equiseti* which is considered to be a dangerous pathogen of rye seedlings as well as of roots and stem bases of wheat and oats (Majchrzak et al. 2008; Kiecana et al. 2008, 2009). This fungus has also contributed to damaging the stem base in triticale (Pląskowska et al. 2010).

The species *F. sporotrichioides* was isolated from the infected roots and lower internodes of rye stems under the growing conditions in question; it has been observed with varying frequency on seedlings, roots and stem bases of grains grown in Poland (Kiecana et al. 2008, 2009; Pląskowska et al. 2010).

In addition to *Fusarium* spp., the species *B. sorokiniana* and *R. solani* proved to be pathogens damaging rye roots and stem bases during the 2001-2005 study period, likewise in the case of spring barley and oats (Łacicowa et al. 1997; Kiecana et al. 2008; Fernandez et al. 2009).

Bipolaris sorokiniana plays an important role in causing stem base diseases in cereals grown across the world (Łacicowa et al. 1997; Almgren et al. 1999; Strausbaugh et al. 2004; Kiecana et al. 2008, 2009; Fernandez et al. 2009). The pathogenicity of this species is associated with the production of secondary metabolites that show phytotoxic properties, e.g. prehelminthosporal which reduces the efficiency of the hydrogen and calcium pumps in the plant cell (Olbe et al. 1995). Disturbances in the functioning of the cytoplasmic cell membranes are also a response of the plant to metabolites produced by *B. sorokiniana* (Wiśniewska et al. 1998).

The species *Aureobasidium pullulans*, which was frequently isolated from rye roots and stem bases in the study of by Łacicowa et al. (1997), proved to be a weak pathogen which, occurring simultaneously with *B. sorokiniana*, impaired spring barley emergence and increased the severity of disease symptoms in the plants.

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Grzyby porażające korzenie i podstawę źdźbła żyta ozimego (*Secale cereale* L.) uprawianego w województwie lubelskim

Streszczenie

Badania przeprowadzono w latach 2001-2005 na 10 plantacjach produkcyjnych żyta ozimego zlokalizowanych w województwie lubelskim. Udział źdźbeł żyta ozimego z objawami nekrozy na dolnych międzywęźlach wahał się od 32,5% do 71% w 2001, od 38% do 68% w 2002, od 21,5% do 56,5% w 2003, od 35% do 70% w 2004 i od 36% do 88% w 2005. Średnie wskaźniki chorobowe wahały się od 7,5 do 46,75 i różniły się istotnie. Wyniki analizy mykologicznej wykazały, że głównymi patogenami porażającymi korzenie i podstawę źdźbła żyta były gatunki *Fusarium avenaceum* (24% ogółu wyosobnień) i *F. culmorum* (25% wszystkich izolatów).