

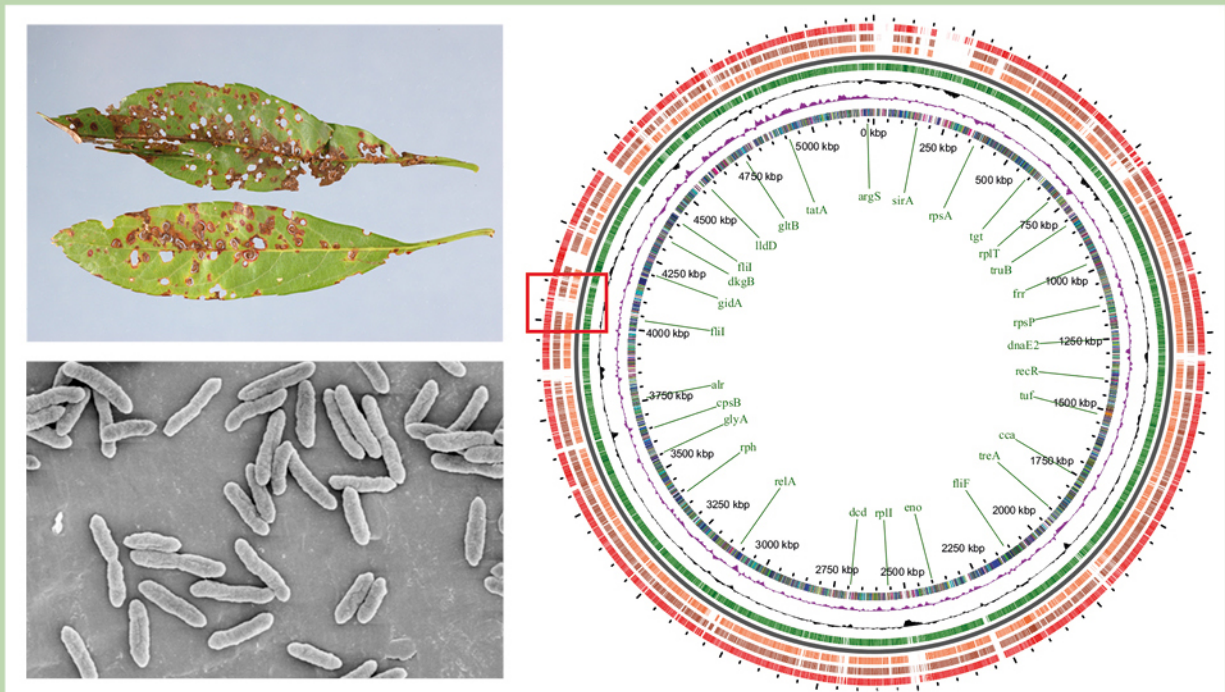


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## BIODIVERSITY OF THE *FUSARIUM* FUNGI CAUSING ROOT ROT OF WINTER CEREALS IN BELARUS

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*Fusarium* fungi are the main causal agents of root rot of winter cereals in Belarus. As many as 12 different species were identified, with occurrence being dependent on the cereal host species and weather conditions during the growing season. Lack of precipitation from April to July led to decreasing fungal biodiversity. *Fusarium* pathogen complex on wheat roots was formed by *F. culmorum*, *F. avenaceum*, *F. equiseti*, *F. oxysporum*, whereas *F. equiseti* and *F. oxysporum* prevailed on triticale and rye, and *F. solani*, *F. avenaceum* and *F. equiseti* dominated on barley root systems. The infestation of root with *F. oxysporum* increased under dry conditions. In contrast, *F. culmorum* was isolated from root system of wheat and triticale more frequently when rainfall was sufficient. For the first time, *F. cerealis* and *F. tricinctum* were isolated from triticale, and *F. cerealis* from rye only, in the Republic of Belarus.

**Keywords:** *Fusarium* root rot, species composition, pathogen complex, occurrence frequency, weather conditions

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### Introduction

Root rot is one of the most widespread diseases of winter cereal crops in many regions of the world (Fernandez et al., 2007; Tunali et al., 2008; Grigoryev, 2012b; Moya-Elizondo et al., 2015; Kolomiets, 2016; Xu et al., 2018). Depending on disease aetiology, several kinds of the root rot are distinguished, with differing prevalence in different regions of cultivation (Paulitz et al., 2002; Cromley et al., 2006; Grigoryev, 2012b). Yield losses due to the root rot average 9% , but in years of intensive disease development, they can be as high as to 35% (Paulitz et al., 2002; Poole et al., 2013).

The *Fusarium* root rot is the most widespread and harmful under conditions typical of Belarus (Buga et al., 2000; Sklimenok, 2015; Krupenko et al., 2016). The fungus *Bipolaris sorokiniana* (Sacc.) Shoemaker in certain years also joins the pathogen complex of fungi causing the disease. Its prevalence may reach 30% at the period of earing, followed by a significant decrease due to antagonistic interactions among the fungi (Sklimenok, 2015).

The importance of studies of fungi in the genus *Fusarium* Link is acknowledged around the world because of their

presence in numerous agricultural crops, high harmfulness, and production of the mycotoxins which are dangerous to the health of humans and domestic animals (Zinedine et al., 2007; Marin et al., 2010; Pestka, 2010). In cereal crops, this problem is augmented by recent changes of species composition of the plant pathogenic fungi, which can be linked to climate change (Tunali et al., 2008; Moya-Elizondo et al., 2011).

Investigation of species composition of the fungi causing root rot in winter cereals is performed in Belarus since the end of the XX century (Buga et al., 2000). Most studies focused on the winter wheat, which is the most widely grown crop in the republic. Yet, during the last 20 years, there have been changes in species composition for the pathogens, with previously infrequent species becoming the prevailing ones (Sklimenok, 2015; Krupenko, Kryzhanovskaya, 2017). To forecast the harmfulness of the root rot fungi and to develop measures to protect crops from the disease, species composition of its causative agents needs to be thoroughly examined.

### Materials and Methods

To analyze species composition of the root rot pathogens of winter cereal crops, surveys of wheat, triticale, rye, and barley stands were performed in 2018–19. The territory of Belarus extends for 650 km from west to east; therefore, each crop was sampled in western, central and eastern parts of the country to account for possible geographic variation. Both plants with and without root rot symptoms (10 plants per sample) were collected at the stage of the wax ripeness (Table 1).

The samples were dried and stored refrigerated. The fungi of the genus *Fusarium* were isolated as follows. The roots were rinsed with tap water for one hour, cut into pieces 1–1.5

cm long, disinfected by immersion into 1% solution of sodium hypochlorite for 15–20 sec, rinsed twice with sterile distilled water, and placed between layers of sterile blot paper. Then the samples were placed in Petri dishes onto potato sucrose agar (PSA) with addition of Triton X-100 and 5% streptomycin. The dishes were incubated for 10 days at 22 °C, then the grown colonies of the *Fusarium* fungi were transferred to PSA in glass tubes. Micro- and macromorphology were used for species identification according to Gerlach & Nirenberg (1982).

Incidence level of each species was estimated as a percentage of its isolates among the total number of the

**Table 1.** Information on the origin of the samples of winter cereals and number of *Fusarium* fungi included in present study

Geographic zone of Belarus	Region	District	Crop	Number of <i>Fusarium</i> isolates	
				2018	2019
Eastern	Mogilev	Gorki	wheat	117	3
			triticale	40	13
			rye	23	3
			barley	-	10
Central	Minsk	Minsk	wheat	198	1
			triticale	11	7
			rye	16	17
Western	Grodno	Schuchin	wheat	45	2
			triticale	35	7
	Brest	Kobrin	rye	29	10

*Fusarium* fungi colonies in each sample and averaged estimates were calculated. Species diversity was evaluated using the Shannon's index according to the formula

$$H_w = -\sum p_i * \ln(p_i),$$

where  $p_i$  is the frequency of the given phenotype in the population (Kolmer et al., 2003).

### Results

In April–July of 2018, the air temperature and precipitation levels were elevated. In 2019, the air temperature at the whole Belarus territory exceeded the average perennial values while the precipitation was below normal, especially in the western part of the country (Grodno region), with a total precipitation during four months of 167.9 mm (vs normal 264.0 mm). As a result, in 2018 the conditions favored increased diversity of the *Fusarium* fungi, as can be seen from rather high Shannon's index values: 2.83 (rye), 2.92 (triticale), and 4.19 (wheat). In 2019, deficiency of precipitation resulted in lower fungal diversity: 0.22 (wheat), 0.81 (barley), 1.91 (triticale), and 2.02 (rye).

The core group of the root rot pathogen complex was made up by *F. culmorum* and *F. avenaceum*, as well as by *F. equiseti*

and *F. oxysporum* in winter wheat; *F. equiseti* and *F. oxysporum* in triticale and rye; and *F. solani*, *F. avenaceum*, and *F. equiseti* in barley (Table 2).

Incidence levels of *F. avenaceum* reached 33.3%, 20.0%, 9.5% and 4.7% of the total fungal complex in wheat, barley, rye and triticale stands, respectively. The highest level of *F. culmorum* was in wheat (up to 30.9%) and triticale (up to 16.9%) under the conditions of sufficient humidity.

Besides the aforementioned species, *F. cerealis*, *F. graminearum*, *F. poae* (Peck) Wollenw., *F. sambucinum* Fuckel, *F. semitectum* Berk. & Ravenel, *F. sporotrichioides* Sherb. and *F. tricinctum* (Corda) Sacc. were also found in root system, with incidence levels depending upon the crop.

**Table 2.** Species composition of the *Fusarium* fungi on the root system of winter cereals

Species of <i>Fusarium</i>	Incidence level, %						
	wheat		triticale		rye		barley
	2018 г.	2019 г.	2018 г.	2019 г.	2018 г.	2019 г.	2019 г.
<i>F. avenaceum</i>	18.5±5.9	33.3±33.3	6.1±6.1	9.5±9.5	4.7±0.9	2.0±2.0	20.0
<i>F. cerealis</i>	4.2±4.2	0.0	0.9±0.9	0.0	2.1±2.1	2.0±2.0	0.0
<i>F. culmorum</i>	30.9±11.4	0.0	16.9±14.3	9.5±9.5	5.3±2.7	10.0±10.0	0.0
<i>F. equiseti</i>	5.8±3.2	33.3±33.3	30.2±18.3	34.4±10.1	32.1±16.1	18.6±15.8	20.0
<i>F. graminearum</i>	0.2±0.2	0.0	0.0	0.0	1.4±1.4	0.0	0.0
<i>F. oxysporum</i>	6.9±3.0	16.7±16.7	36.0±15.8	28.9±21.7	28.6±5.9	49.8±25.2	0.0
<i>F. poae</i>	0.0	0.0	1.7±1.7	0.0	2.6±1.3	0.0	0.0
<i>F. sambucinum</i>	0.0	0.0	1.7±1.7	0.0	0.0	0.0	0.0
<i>F. semitectum</i>	0.0	0.0	0.0	0.0	2.1±2.1	2.0	0.0
<i>F. solani</i>	2.3±1.3	16.7±16.7	0.8±0.8	0.0	4.7±0.9	0.0	50.0
<i>F. sporotrichioides</i>	6.0±3.9	0.0	4.0±2.7	7.7±7.7	14.4±8.5	13.7±13.7	0.0
<i>F. tricinctum</i>	0.0	0.0	0.0	0.0	2.1±2.1	2.0±2.0	0.0
<i>Fusarium</i> spp.*	25.3±1.9	0.0	1.8±0.9	9.9±9.9	0.0	0.0	10.0

\* unidentified species from different sections

### Discussion

Results of this research indicate that formation of *Fusarium* root rot pathogens' complex is largely influenced by weather conditions during the vegetation period. Higher precipitation level, resulted in higher species diversity of the *Fusarium* fungi. The weather may also promote certain species over other species. Our study has confirmed that dry and hot years

are beneficial for *F. oxysporum*, while cold and humid years favor to *F. culmorum* (Grigoryev, 2012a; Poole et al., 2013).

Species diversity of fungi is known to vary greatly even within a limited area (Xu, Nicholson, 2009; Khemir et al., 2020), leading to considerable changes in the prevailing species range, which determine harmfulness of plant diseases. This explains why monitoring of the composition of root rot

fungi species complexes and identifying dominant species are essential for improving plant protection. In Belarus, the species composition of causative agents of Fusarium root rot is changing. In the beginning of the first decade of XXI century, main root rot pathogens of winter wheat, rye and triticale were *F. oxysporum*, *F. culmorum* and *F. sporotrichiella* (Buga et al., 2000). Ten years after, *F. oxysporum* was still prevailing but importance of *F. avenaceum* and *F. equiseti* has also increased (Sklimenok, 2015) and in certain years, *F. culmorum* and *F. solani* were frequent (Sklimenok, 2015; Krupenko, Kryzhanovskaya, 2017). In the present study conducted in

2018–2019, the dominant species were *F. avenaceum* and *F. culmorum*, as well as *F. equiseti* and *F. oxysporum* (under conditions of rainfall deficiency). Differences in the prevailing species of *Fusarium* were also found between the winter crops. In particular, in triticale and rye the dominating species of the pathogens' complex were *F. equiseti* and *F. oxysporum*, while in barley those were *F. solani*, *F. avenaceum*, and *F. equiseti*.

Two pathogen species have been isolated for the first time in Belarus from the root system of winter triticale (*F. cerealis* and *F. tricinctum*) and rye (*F. cerealis*), which were previously reported in winter wheat only (Sklimenok, 2015).

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## ВИДОВОЕ РАЗНООБРАЗИЕ ГРИБОВ РОДА *FUSARIUM*, ВЫЗЫВАЮЩИХ КОРНЕВУЮ ГНИЛЬ ОЗИМЫХ ЗЕРНОВЫХ КУЛЬТУР В БЕЛАРУСИ

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Представлены результаты изучения видового состава грибов рода *Fusarium*, являющихся основными возбудителями корневой гнили озимых зерновых культур (пшеница, рожь, тритикале, ячмень). Выявлено не менее 12 разных видов, частота встречаемости которых варьировала в зависимости от вида зерновой культуры и погодных условий в период сезона вегетации. Основу комплекса патогенов грибов-возбудителей корневой гнили на озимой пшенице (в порядке уменьшения частоты встречаемости) составили виды *F. culmorum*, *F. avenaceum*, *F. equiseti*, *F. oxysporum*. На тритикале и ржи преобладали грибы *F. equiseti* and *F. oxysporum*, на ячмене – *F. solani*, *F. avenaceum* and *F. equiseti*. Установлено, что в условиях дефицита осадков видовое разнообразие снижалось. Частота встречаемости гриба *F. culmorum* на пшенице и тритикале была выше в год с избыточным увлажнением, тогда как *F. equiseti* and *F. oxysporum* на озимых тритикале и ржи она была высокой независимо от года, при этом доля последнего увеличивалась в условиях недостатка осадков. Впервые в Беларуси из корневой системы озимых тритикале и ржи выделен *F. cerealis*, а также *F. tricinctum* – из озимой ржи.

**Ключевые слова:** фузариозная корневая гниль, видовой состав, комплекс патогенов, частота встречаемости, гидротермические условия

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