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Fusarium head blight (FHB) and *Fusarium* spp. on grain of spring wheat cultivars grown in Poland

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Abstract: Eighteen spring wheat cultivars, recommended for commercial production in northern Poland, were assessed for Fusarium head blight (FHB) in natural non-epidemic conditions, from 2011 to 2013. Assessment was based on FHB incidence (proportion of heads with symptoms), disease severity (DS; proportion of bleached spikelets per head), proportion of *Fusarium* damaged kernels (FDK), and spectrum of *Fusarium* spp. colonising the kernels. Fusarium head blight incidence and DS often differed significantly among cultivars and years. There was a strong positive correlation between FHB incidence and DS. Fusarium head blight incidence and DS were not correlated with the June–July temperatures, and were only occasionally correlated with the total June–July rainfall. There was a weak positive correlation between FHB incidence and proportion of FDK. There was a strong positive correlation between DS and proportion of FDK. The cultivar affected colonisation of kernels by *Fusarium* spp. *Fusarium poae* was the FHB pathogen isolated most often. *Fusarium poae* colonised 6.0% of the kernels, on average, but up to 12.0% on individual cultivars. Other *Fusarium* species were less frequent: *F. avenaceum* in 5.6% of kernels, *F. culmorum* in 5.3%, *F. tricinctum* in 2.8%, *F. graminearum* in 1.5%, and *F. sporotrichioides* in 1.2%. *Fusarium equiseti* occurred sporadically. The importance of *F. poae* in the FHB complex is emphasised. All cultivars expressed 'moderate FHB resistance' if evaluated according to FHB incidence. Cultivars Arabella, Izera, Kandela, Monsun, Ostka Smolicka, and Struna expressed 'moderate susceptibility', and Bombona, Hewilla, Katoda, KWS Torridon, Łagwa, Nawra, Parabola, Radocha, SMH 87, Trappe, Tybalt, and Waluta expressed 'susceptibility' if evaluated by the proportion of FDK. Cultivars differed within the 'moderately resistant', 'moderately susceptible', and 'susceptible' categories. Cultivars Arabella, Izera, Kandela, Monsun, Ostka Smolicka, and Struna were the most promising and their resistance traits may be useful in FHB management.

Key words: cultivar, *Fusarium*, Fusarium head blight (FHB), spring wheat, susceptibility

Introduction

Spring wheat is the second most valuable cereal after winter wheat. It is grown in Poland on 380 000 ha, particularly in the north and in Lower Silesia (southern Poland). The area has increased recently because of losses occurring in winter crops. Spring wheat's advantages make it popular in large-scale and commercial production, and locally on ecological farms.

Spring wheat cultivars bred in Europe are characterised by short, lodging-resistant stalks (usually decreasing FHB, which can be more severe in crops that lodge), higher yields, and high efficiency in mineral nutrition. The quality of the grain depends on the genetic features of the cultivar, weather conditions, and agricultural management. Spring wheat may have less yield than its winter equivalent. Instead, the popularity of spring wheat results from its agricultural value. Spring wheat has a shorter period of growth and of exposure to pathogens, pests, and weeds. All of these features decrease cultivation costs.

Fusarium head blight (FHB) is one of the most important cereal diseases. It has emerged as a major threat to wheat and barley crops around the world. Fusarium head blight contributes to loss of grain yield and quality due to colonisation by *Fusarium* fungi and contamination with mycotoxins (Siuda *et al.* 2010; Grabowski *et al*. 2012a, b; Gromadzka *et al*. 2012).

The prevalence of disease depends on agronomic practices, effectiveness of fungicides used, and host resistance. In Poland, FHB has been observed every year with differences in incidence. In some years, incidence was low (< 1% of heads colonized). Recently, FHB severity has increased, and has been seen each year on approximately 70% of wheat fields (Wakuliński *et al*. 1991; Lenc *et al.* 2011; Sadowski *et al.* 2011; Lenc 2015).

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Seventeen species of *Fusarium*, with different climatic requirements and genetic and environmental adaptations, contribute to disease (Parry *et al*. 1995; Stępień and Chełkowski 2010; Wiśniewska *et al.* 2014). *Fusarium graminearum* Schwabe [*Gibberella zeae* (Schwein.) Petch] is the predominant causal agent of FHB in most areas. This fungus is the most virulent worldwide. In Poland, FHB is caused mostly by *F. culmorum* (Wm. G. Sm.) Sacc., *F. avenaceum* (Fr.) Sacc. (*Gibberella avenacea* R.J. Cook) and *F. poae* (Peck) Wollenw. (Wakuliński and Chełkowski 1993; Lenc 2015). The contribution of particular species to FHB incidence depends on habitat and weather conditions.

Deoxynivalenol (DON) and zearalenone are the most common mycotoxins associated with FHB They pose a serious threat to human and domestic animal health. Crop sequence and tillage (which incorporates crop residues into the soil) have been shown to affect the incidence of FHB. In recent years, decreased tillage is thought to have contributed to regional epidemics by increasing the levels of inoculum available for infection. Additionally, disease is favored by extended periods of moderately high temperature (15–30°C), high moisture (relative humidity > 90%), and frequent rainfall (De Wolf *et al*. 2003; Lemmens *et al*. 2004; Xu *et al*. 2008; Sadowski *et al*. 2011; Lenc 2015).

Extensive research aimed at controlling FHB has focused on the development of wheat cultivars resistant to *Fusarium* spp. and the use of such resistant cultivars in integrated management systems. The development of resistant cultivars is the most effective, economic, and environmentally safe way to control FHB in wheat. Thousands of plant lines are subjected to artificial inoculation with *Fusarium* spp. (mostly with *F. graminearum*). Those lines having reduced fungal growth and low levels of seed contamination with DON, are selected and advanced in further breeding trials. Quantitative Trait Loci (QTL) composed of one or more genes, such as *Fhb*1 derived from the Chinese wheat cultivar Sumai 3, have been identified and widely used in wheat breeding (Steiner *et al.* 2004; Góral 2005; Zhang *et al*. 2008; Kubo *et al*. 2013).

Two main types of resistance to FHB are widely accepted: Type I, resistance to initial infection; and Type II, resistance to fungal spread within the spike (Schroeder and Christensen 1963). Three other types of resistance were reported by Mesterhazy *et al.* (1999): Type III, resistance to DON accumulation; Type IV, resistance to kernel infection; Type V, tolerance. Infection and disease development can be affected by both active (i.e. physiological processes) and passive (i.e. plant height, spike architecture, flowering date) resistance factors that are difficult to separate (Crute *et al.* 1985; Mesterhazy 1995; Buerstmayr *et al.* 2009). Integrated management of FHB in wheat depends on disease forecasting models which help to determine the risk of FHB infection and optimise the agronomic and chemical control of disease. The risk of FHB infection depends partly on cultivar resistance to *Fusarium* spp., which must be considered in commercial production. Genetic variability is essential for the development of FHB resistant cultivars. Sources of FHB resistance/tolerance in spring wheat have so far been introduced from China, Brazil, Europe, and Japan (Fedak *et al.* 2001). There is always a chance of finding further sources of resistance in local cultivars and lines.

The objective of this study was to assess: (i) FHB incidence and disease severity in 18 cultivars of spring wheat grown in a conventional system in northern Poland in the 2011–2013 time period; (ii) effects of weather conditions on FHB incidence and severity; (iii) populations of *Fusarium* fungi involved in FHB, and hence, implications for human and domestic animal mycotoxicoses; (iv) suitability of particular spring wheat cultivars for FHB resistance breeding, commercial production, and requirements of integrated management. The objectives are of particular significance in terms of the European Commission Regulation No. 856/2005 of 6 June 2005 regarding food safety and *Fusarium* toxin concentrations in food and feed.

Materials and Methods

Site description

Seventeen common spring wheat (*Triticum aestivum* L.) cultivars (Arabella, Bombona, Hewilla, Izera, Kandela, Katoda, KWS Torridon, Ławga, Monsun, Nawra, Ostka Smolicka, Parabola, Radocha, Struna, Trappe, Tybalt, and Waluta) from elite groups (A–E), and one durum spring wheat (*T. turgidum* ssp. *durum*) cultivar (SMH 87) were grown in a conventional system in experimental fields at the Experimental Station for Variety Testing in Lisewo Malborskie, northern Poland (54° 6′ N, 18° 43′ E) from 2011 to 2013. Not all cultivars were grown in each of the study years. The experiment was established with four replicates in a randomised block design in brown soil of class I quality. The chemical characteristics of the soil at Lisewo (2011–2013) are presented in table 1. The previous crop in each year was the sugar beet (*Beta vulgaris* L.). Sowing was done on 8–10 April at 450–500 grains · m^{-2}. Fertiliser application each year was: 140 kg NH_4^+ · ha^{-1}, 70 kg P_2O_5 · ha^{-1} and 100 kg K_2O · ha^{-1}. Fungicidal seed treatments, and herbicide and insecticide sprays were applied each year (Table 2). Grain was harvested each year on 27–30 August.

The average 2011–2013 temperatures in June were 15.5–17.8°C, and in July 18.3–18.6°C, with the highest temperatures in July 2012 (Table 3). Total rainfall in June was from 36.2 to 125.1 mm, and in July from 102.3 to 170.3 mm, with the most rain in July 2013. The number of days with rain was in the range of 8–18 in June, and 12–20 in July. Although temperatures were moderate in June–July 2011–2013, the high rainfall in June 2012 and July 2011–2013, and the high number of days with rainfall, particularly in July 2011–2013, meant that weather conditions were generally favorable for FHB development.

Collection of samples and disease assessment

Each year (2011–2013), 50 heads of wheat from each cultivar were collected at late milk to early dough development stages (GS 77-83; Zadoks *et al*. 1974). These wheat heads were collected from randomly chosen plants along a diagonal transect across each of the four replicate plots. Fusarium head blight incidence in heads, and disease se-

Table 1. Chemical characteristics of soil at Lisewo, in 2011–2013

Soil characteristics	2011	2012	2013
pH in KCl	5.9	5.8	6.2
Extractable soil phosphorus [mg · kg^{-1}][a]	9.9	24.4	18.2
Extractable soil potassium [mg · kg^{-1}][b]	17.8	20.0	25.1
Extractable soil magnesium [mg · kg^{-1}][b]	12.6	12.4	13.5

[a]analysed with the Egner-Riehm method
[b]analysed with the Schachtschabel method

Table 2. Pesticides used in spring wheat production at Lisewo, in 2011–2013

Seed treatment	2011	2012	2013
Fungicide	Sarfun T 65 DS[1] (200 g · 100 kg^{-1})	Sarfun T 65 DS (200 g · 100 kg^{-1})	Zaprawa T 75DS/WS[2] (200 g · 100 kg^{-1})
Herbicide	Gold 450 EC[3] + Gallaper 200 EC[4] (1.0 l + 0.8 l ha^{-1})	Granstar Ultra[5] + Tomigan 250 EC[6] (40 g + 0.5 l ha^{-1})	Granstar Ultra + Hurler 200 EC[7] (40 g + 0.4 l ha^{-1})
Insecticide	Karate Zeon 050CS[8] (0.12 l · ha^{-1})	Karate Zeon 050CS (0.1 l · ha^{-1})	Wojownik 050SC[9] (0.1 l · ha^{-1})

[1]Carbendazim 20% + thiuram 45%; [2]Thiuram 75%; [3]Terbuthrine + metolachlor; [4]Fluoroxypyr; [5]Tribenuron methyl 50% + trisodium phosphate dodecahydrate 20%; [6]Fluroxypyr 25%; [7]Fluroxypyr meptyl 20%; [8]Lambda-cyhalothrin + 1,2-benzisothiazolin-3-one; [9]Lambda cyhalothrin 5%

Table 3. Temperature and rainfall during the flowering and ripening stages of spring wheat growth at Lisewo, in 2011–2013 (according to Experimental Station for Variety Testing in Lisewo Malborskie)

Month	10-day period	Average temperature [°C]			Total rainfall [mm]			Number of days with rainfall		
		2011	2012	2013	2011	2012	2013	2011	2012	2013
June	I	19.1	13.0	15.8	9.1	16.7	5.6	3	6	2
	II	16.5	16.9	18.7	18.1	24.9	0.5	4	6	2
	III	17.7	16.8	17.8	11.7	83.5	30.1	2	6	4
	average	17.8	15.5	17.4	38.9	125.1	36.2	9	18	8
July	I	17.6	20.0	18.2	47.0	60.6	88.8	6	8	4
	II	19.4	16.2	17.4	45.9	23.8	38.2	6	8	4
	III	18.2	19.6	19.4	15.2	17.9	43.3	6	4	4
	average	18.4	18.6	18.3	108.1	102.3	170.3	18	20	12

verity, were evaluated visually on 200 heads from each cultivar. Fusarium head blight incidence was determined as the proportion (%) of heads with symptoms. Disease severity (DS), i.e. the extent of head damage (%), was determined as the proportion of bleached spikelets per head, based on a 1–9 scale: 1 – no symptoms; 2 – < 5% of bleached spikelets; 3 – 5–15%; 4 – 16–25%; 5 – 26–45%; 6 – 46–65%; 7 – 66–85%; 8 – 86–95%; 9 – 96–100% (Miedaner and Perkowski 1996).

Colonisation of wheat kernels by fungi

Mycological analysis of 400 (4 × 100) wheat kernels collected randomly during harvest from the four plots of each cultivar was performed each of the study years. In the laboratory, the kernels were rinsed for 45 min in running water, disinfected in 1% NaOCl solution for 2.5 min, rinsed three times for 10 min in sterile distilled water, and placed on Potato Dextrose Agar (PDA; boiled and sieved white potatoes 400 g · l^{-1}, agar 20 g · l^{-1}, streptomycin 50 mg · l^{-1}, pH = 7) in Petri dishes. Fungi were incubated for 7–10 days at 20°C in a day-night cycle. All colonies on each plate were then examined macro- and microscopically and distinguished on the basis of colour, growth rate, hyphal characteristics, and sporulation. Colonies of each species were counted and representative fungi were identified by morphotyping on PDA and Synthetic Nutrient Agar (SNA; KH_2PO_4 1 g · l^{-1}, KNO_3 1 g · l^{-1}, $MgSO_4 \cdot 7H_2O$ 0.5 g · l^{-1}, KCl 0.5 g · l^{-1}, glucose 0.2 g · l^{-1}, sucrose 0.2 g · l^{-1}) using Booth (1971), Kwaśna *et al.* (1991). Proportions (%) of *Fusarium*-colonised kernels (FDK) were calculated.

Evaluation of cultivar response to *Fusarium* infection

The response of each cultivar was determined from FHB incidence and the proportion of FDK. For response based on the FHB incidences (proportion of heads with symptoms), the following scale was used: 0 – immune; 1–5% – resistant; 5–25% – moderately resistant; 25–50% – moderately susceptible; 50–75% – susceptible; > 75% – very susceptible. For response based on the proportion of FDK, the scale was as follows: 0 – immune; 1–8% – resistant; 9–11% – moderately resistant; 12–20% – moderately susceptible; 21–50% – susceptible; > 50% – very susceptible.

Statistical analysis

The statistical significance of differences in FHB incidence and in DS on different cultivars was tested using one-way analysis of variance (ANOVA, $p \leq 0.05$) and Tukey's *post hoc* test, (FR-ANALWAR software). Percentage values were transformed into Bliss degrees before statistical analysis. The statistical significance of differences in the number of FDK and the statistical significance of differences in the number of kernels colonised by individual *Fusarium* species were determined using χ^2 tests. The null hypothesis assumed that wheat from different systems had the same number of kernels colonised by *Fusarium* spp. Pearson's correlation coefficient was applied to analyse the relationships between FHB incidence, DS, proportion of FDK, yield, temperature, and rainfall.

Results

Effects of cultivar on disease

Fusarium head blight (FHB) incidence and DS differed among cultivars and years, often significantly (Table 4). Differences were not usually consistent. In 2011, there was no FHB on cvs Kandela, Ługwa or Ostka Smolicka, and the most FHB was on cvs Parabola (11.5%) and Tybalt (9.5%). There was more FHB in 2012 and 2013. In 2012, the least FHB incidence occurred on cv. Trappe (7%) and the most on cvs Bombona, Izera, Kandela, Katoda, KWS Torridon, Tybalt, and Waluta (10.5–14%). In 2013, a low FHB incidence (6.5–10%) was observed on a few cultivars including Arabella (6.5%) and most occurred again on cv. Tybal (25.5%). There was strong positive correlation between FHB incidence and DS ($r = -0.925$, $p \leq 0.001$).

Effects of weather on disease

In 2011, 2012, and 2013, FHB incidence and DS were not correlated with the June–July temperatures. Fusarium head blight incidence was not correlated with total rainfall. Only DS was correlated, at a low level, with July rainfall ($r = 0.516$–0.657, $p \leq 0.001$). June rainfall was least in 2011 and 2013 but was associated with lower average FHB and DS values only in 2011.

Colonisation of kernels by *Fusarium* spp.

Cultivar affected the colonisation of kernels by *Fusarium* spp. There were significant differences in proportions of FDK in cultivars in each of the years of the study (Table 5). Averaged over three years, FDK ranged from 13.0% (cv. Izera) to 35.3% (cv. Tybalt). In cvs Katoda, KWS Torridon, Monsun, Parabola, Radocha, SMH 87, Struna, Trappe, and Tybalt, the high average (2011–2013) FDK (20.0–35.3%) was associated with a high average FHB incidence (10.0–16.3%). In cvs Arabella, Izera, and Kandela, the lower average (2011–2013) FDK (13.0–16.8%) was associated with a lower average FHB incidence (6.7–9.5%). The average FDK of all cultivars was similar in all the years of the study. There was a weak, but significant, positive correlation between proportion of FDK and FHB incidence ($r = 0.594$, $p \leq 0.001$) and a stronger positive correlation between proportion of FDK and DS ($r = 0.733$, $p \leq 0.001$). There was a weak, but significant, negative correlation between total rainfall in July and the proportion of FDK ($r = -0.636$, $p \leq 0.001$).

Fungal species in kernels

Cultivar significantly affected colonisation of kernels by individual *Fusarium* species (χ^2 test, $p \leq 0.05$). *Fusarium poae* was the *Fusarium* species isolated most often (Table 6). It was frequent each year, on each cultivar. It colonised 6.0% of the kernels, on average, but up to 12.0% on cv. Tybalt. Other *Fusarium* species occurred less frequently: *F. avenaceum* (*G. avenacea*) in 5.6% of kernels, on average, *F. culmorum* in 5.3%, *F. tricinctum* (*G. tricincta*) in 2.8%, *F. graminearum* (*G. zeae*) in 1.5%, and *F. sporotrichioides* in 1.2% of kernels. *Fusarium equiseti* (*G. intricans*) was the rarest. Wheat kernels were also colonised by other fungi; the most common were *Alternaria alternata*, *Arthrinium phaeospermum*, and *Epicoccum nigrum.*

Yield

The total grain yield or thousand-kernel weight (TKW) was not affected by FHB incidence. There was no correlation between average yield of individual cultivars or average TKW in 2011–2013, and average FHB incidence, DS or proportion of FDK. Average grain yield of individual cultivars in 2011–2013 ranged from 6.09 $t \cdot ha^{-1}$ (cv. SMH 87) to 9.41 $t \cdot ha^{-1}$ (cv. Trappe) (Table 7). The range of the average TKW was from 44.8 g (cv. Trappe) to 56.0 g (cv. Parabola). There was a weak, but significant, negative correlation between average yield of individual cultivars in 2011–2013 and TKW ($r = -0.639$, $p \leq 0.001$). The highest yields, in cvs Trappe (9.41 $t \cdot ha^{-1}$) and KWS Torridon (9.32 $t \cdot ha^{-1}$), were associated with the lowest TKW (44.8 g and 45.9 g, respectively) (Table 8). The lowest yield, in cv. SMH 87 (6.09 $t \cdot ha^{-1}$), was associated with high TKW (53.2 g), and high FHB incidence (13.5%) and DS (5.2%).

Cultivar response to *Fusarium* infection

All cultivars expressed 'moderate resistance' when assessed according to the FHB incidence (Table 8). Cultivars

Table 4. Fusarium head blight (FHB) incidence and disease severity (DS) on different cultivars of spring wheat at Lisewo, in 2011–2013

Cultivar	FHB [%]				DS [%]			
	2011	2012	2013	2011–2013	2011	2012	2013	2011–2013
Arabella	–	10.0^{a,b,c,d}	6.5^{f}	[1]8.3^{b,c,d,e}	–	4.1^{b,c,d}	2.3^{i}	[1]3.2^{c,d,e,f,g}
Bombona	5.0^{b,c}	12.5^{a,b}	10.0^{c,d,e,f}	9.2^{b,c,d}	2.3^{a,b,c}	5.5^{a,b}	4.1^{e,f,g,h}	4.0^{c,d,e,f,g}
Hewilla	4.0^{c}	8.0^{c,d}	10.0^{c,d,e,f}	7.3^{c,d,e,f}	2.2^{b,c}	3.4^{c,d}	3.7^{f,g,h,i}	3.1^{d,e,f,g}
Izera	–	11.5^{a,b,c}	7.5^{e,f}	[1]9.5^{a,b,c}	–	4.6^{a,b,c,d}	2.6^{h,i}	[1]3.6^{c,d,e,f,g}
Kandela	0.0^{d}	11.0^{a,b,c}	9.0^{d,e,f}	6.7^{d,e,f}	0.0^{c}	4.7^{a,b,c}	3.4^{g,h,i}	2.7^{f,g}
Katoda	7.5^{a,b,c}	11.0^{a,b,c}	13.0^{b,c,d}	10.5^{a,b,c}	4.2^{a,b}	4.6^{a,b,c,d}	4.7^{d,e,f,g}	4.5^{b,c,d,e,f}
KWS Torridon	–	10.5^{a,b,c,d}	13.0^{b,c,d}	[1]11.8^{a,b,c}	–	4.4^{a,b,c,d}	5.3^{c,d,e}	[1]4.9^{a,b,c,d}
Łagwa	0.0^{d}	7.5^{c,d}	8.5^{d,e,f}	5.3^{f}	0.0^{c}	5.5^{a,b}	3.0^{h,i}	2.8^{e,f,g}
Monsun	–	–	11.0^{b,c,d,e,f}	[2]11.0^{a,b,c}	–	–	3.6^{f,g,h,i}	[2]3.6^{c,d,e,f,g}
Nawra	9.0^{a,b}	–	–	[2]9.0^{b,c,d}	5.0^{a,b}	–	–	[2]5.0^{a,b,c,d}
Ostka Smolicka	0.0^{d}	8.0^{c,d}	9.0^{d,e,f}	5.7^{e,f}	0.0^{c}	3.2^{c,d}	3.4^{g,h,i}	2.2^{g}
Parabola	11.5^{a}	–	15.5^{b,c}	[1]13.5^{a,b}	6.4^{a}	–	6.2^{b,c}	[1]6.3^{a,b}
Radocha	–	9.0^{b,c,d}	14.0^{b,c,d}	[1]11.5^{a,b,c}	–	3.8^{b,c,d}	5.8^{b,c,d}	[1]4.8^{b,c,d,e}
SMH 87	–	–	13.5^{b,c,d}	[2]13.5^{a,b}	–	–	5.2^{c,d,e}	[2]5.2^{a,b,c}
Struna	–	–	10.0^{c,d,e,f}	[2]10.0^{a,b,c}	–	–	4.0^{e,f,g,h}	[2]4.0^{c,d,e,f,g}
Trappe	–	7.0^{d}	16.5^{b}	[1]11.8^{a,b,c}	–	2.9^{d}	7.0^{b}	[1]5.0^{a,b,c,d}
Tybalt	9.5^{a}	14.0^{a}	25.5^{a}	16.3^{a}	5.1^{a,b}	6.1^{a}	9.6^{a}	6.9^{a}
Waluta	4.0^{c}	11.0^{a,b,c}	12.0^{b,c,d,e}	9.0^{b,c,d}	2.1^{b,c}	4.6^{a,b,c,d}	5.0^{c,d,e,f}	3.9^{c,d,e,f,g}
Average	5.1	10.1	12.0	10.0	2.7	4.4	4.6	4.2
Least significant difference (LSD) at p = 0.05					4.13	1.78	1.57	2.02

[1]data from two years
[2]data from one year
A different letters in a column indicates significant difference according to one-way ANOVA at $p \leq 0.05$

Table 5. Proportion (%) of *Fusarium* damaged kernels (FDK) in different cultivars of spring wheat at Lisewo, in 2011–2013

Cultivar	2011	2012	2013	2011–2013
Arabella	–	17.0^{f}	13.0^{g,h}	[1]15.0^{i,j}
Bombona	22.0^{c,d}	22.0^{c,d,e,f}	21.0^{c,d,e,f}	21.7^{e,f,g}
Hewilla	31.0^{a,b}	20.0^{e,f}	22.0^{b,c,d,e,f}	24.3^{c,d,e}
Izera	–	16.0^{f}	10.0^{h}	[1]13.0^{j}
Kandela	16.5^{d}	18.0^{e,f}	16.0^{f,g}	16.8^{h,i}
Katoda	35.0^{a}	30.0^{a,b}	27.0^{b,c}	30.7^{b}
KWS Torridon	–	24.0^{b,c,d,e}	22.0^{b,c,d,e,f}	[1]23.0^{d,e,f}
Łagwa	36.0^{a}	21.0^{d,e,f}	10.0^{h}	22.3^{d,e,f,g}
Monsun	–	–	20.0^{d,e,f}	[2]20.0^{f,g,h}
Nawra	25.5^{b,c}	–	–	[2]25.5^{c,d,e}
Ostka Smolicka	26.0^{b,c}	20.0^{e,f}	12.0^{g,h}	19.3^{f,g,h}
Parabola	29.0^{a,b}	–	26.0^{b,c,d}	[1]27.5^{b,c}
Radocha	–	27.0^{a,b,c,d}	25.0^{b,c,d,e}	[1]26.0^{c,d}
SMH 87	–	–	23.0^{b,c,d,e}	[2]23.0^{d,e,f}
Struna	–	–	19.0^{e,f}	[2]19.0^{g,h}
Trappe	–	28.0^{a,b,c}	28.0^{b}	[1]28.0^{b,c}
Tybalt	34.0^{a}	32.0^{a}	40.0^{a}	35.3^{a}
Waluta	29.5^{a,b}	22.0^{c,d,e,f}	22.0^{b,c,d,e,f}	24.5^{c,d,e}
Average	28.5	22.8	20.9	23.1

[1]data from two years
[2]data from one year
A different letters in a column indicates significant difference according to χ^2 test at $p \leq 0.05$

Table 6. Average proportion (%) of kernels colonised by fungi in different cultivars of spring wheat at Lisewo, in 2011–2013

Taxon	Arabella[1]	Bombona	Hewilla	Izera[1]	Kandela	Katoda	KWS Torridon[1]	Łagwa	Monsun[2]	Nawra[2]	Ostka Smolicka	Parabola[1]	Radocha[1]	SMH 87[2]	Struna[2]	Ttrappe[1]	Tybalt	Waluta
Fusarium culmorum (W.G. Sm.) Sacc.	2.5	5.8^{c}	6.3^{bc}	2.5	3.0^{d}	8.2^{ab}	7.0	2.0^{d}	1.0	1.0	2.5	7.0	8.0	9.0	5.0	12.0	10.5^{a}	2.7^{d}
F. poae (Peck) Wollenw.	5.5	7.0^{c}	5.2^{cd}	3.0	4.0^{d}	5.8^{c}	5.5	9.3^{b}	1.0^{e}	9.0^{c}	5.7^{cd}	8.0^{c}	8.0^{c}	2.0^{e}	5.0	8.5^{c}	12.0^{a}	7.0^{c}
F. sporotrichioides Sherb.	1.5	1.2	0.3	0.5	0.3	2.3	0.5	2.0	2.0	4.5	0.8	2.8	0	0	0	1.0	1.3	1.0
Gibberella avenacea R.J. Cook	2.5	3.7^{d}	5.7^{bc}	4.5	5.8^{bc}	7.0^{ab}	4.5	3.7^{d}	11.0^{e}	5.0	5.7^{bc}	4.3^{d}	4.5^{d}	8.0^{ab}	6.0^{bc}	5.0	5.0^{cd}	8.3^{a}
G. intricans Wollenw.	0.5	1.0	0	0	0	0.7	0	0	1.0	0	0	0	0.5	1.0	1.0	0	0.7	0
G. tricincta El-Gholl, McRitchie, Schoult. & Ridings	1.0	2.0	5.3	1.0	2.3	5.0	1.0	4.2	2.0	6.0	4.7	4.0	2.5	2.0	1.0	0.5	3.8	3.2
G. zeae (Schwein.) Petch	1.5	1.0	1.5	1.5	1.3	1.7	4.5	1.2	2.0	0	0	1.5	2.5	0	1.0	1.0	2.0	2.3
Others fungi																		
Acremoniella fusca (Kunze & J.C. Schmidt) Sacc.	1.0	2.0	1.0	0	1.0	1.0	0.5	2.0	0	5.0	3.0	1.0	2.0	0	1.0	3.0	0.3	3.0
Alternaria alternata (Fr.) Keissl.	63.5	62.3	65.0	61.5	60.0	58.7	69.0	61.3	68.0	68.0	60.3	61.0	60.0	66.0	60.0	62.0	55.3	66.0
Arthrinium phaeospermum (Corda) M.B. Ellis	7.5	3.7	9.0	4.5	6.3	4.7	8.5	6.7	9.0	11.0	7.0	11.0	5.5	9.0	8.0	8.0	9.0	7.0
Aspergillus niger Tiegh.	1.0	0	0	0.5	0	0	0	0.7	1.0	0	0	0	0	1.0	1.0	0	0	0.3
Aureobasidium pullulans (de Bary & Löwenthal) G. Arnaud	2.0	0	1.0	1.0	0.7	1.0	1.0	1.0	0	0	1.3	0	1.5	0	0	1.0	0.7	0.7
Bipolaris sorokiniana (Sacc.) Shoemaker	3.5	1.3	3.0	4.0	2.3	0	1.5	1.0	2.0	1.0	0.7	1.5	3.5	3.0	2.0	0.5	0	1.3
Botrytis cinerea Pers.	1.0	1.0	2.0	0.5	2.0	2.3	1.5	1.0	0	1.0	1.0	1.0	1.0	1.0	1.0	2.5	0.7	1.7
Cladosporium herbarum (Pers.) Link	0.5	0.7	0.7	2.5	0.3	0	0	0.7	1.0	0	1.7	0	0.5	2.0	2.0	0	1.0	0.3
Epicoccum nigrum Link	17.5	30.0	27.3	23.5	29.0	30.7	20.0	25.3	19.0	29.0	29.3	22.0	22.0	10.0	22.0	22.5	22.0	22.7
Khuskia oryzae H.J. Huds.	1.0	1.0	1.0	1.5	1.7	1.3	2.5	1.3	1.0	1.0	1.0	0.5		1.0	1.0	0.5	1.7	1.0
Melanospora damnosa (Sacc.) Lindau	3.5	6.0	6.7	2.0	4.7	4.3	2.5	7.3	6.0	7.0	6.7	4.0	3.5	7.0	2.0	3.5	4.0	7.3
Microdochium bolleyi (R. Sprague) de Hoog & Herm.-Nijh.	1.0	1.7	0	0.5	1.0	1.7	0	1.7	0	0	1.3	0	4.0	1.0	1.0	0.5	0.3	1.0
Penicillium spp..	3.5	1.3	3.0	1.5	2.7	1.3	0.5	2.0	0	1.0	4.3	2.0	1.5	0	2.0	1.0	1.0	2.0
Periconia macrospinosa Lefebvre & Aar.G. Johnson	0	0	0.3	0	0	0	0	0	0	0	0.3	0	0	0	0	0	0	0.3
Trichoderma viride Pers.	0	0.3	0	1.0	0.3	0	0	1.0	0	0	0	0	0	0	2.0	0.5	0	0.3
Sterile mycelia	0	0	0	0.5	0.7	1.0	1.0	0	0	1.0	0	0	1.0	0	0	0	0	0

[1]data from two years; [2]data from one year
A different letters in a row indicates significant difference according to χ^2 test at $p \leq 0.05$

Table 7. Grain yield and thousand kernel weight (TKW) in different cultivars of spring wheat at Lisewo, in 2011–2013 (according to the Experimental Station for Variety Testing in Lisewo Malborskie)

Cultivar	Yield [t · ha^{-1}]				TKW [g]			
	2011	2012	2013	average 2011–2013	2011	2012	2013	average 2011–2013
Bombona	8.04	9.29	9.22	8.85	49.0	46.6	42.4	46.0
Hewilla	8.21	8.81	8.77	8.59	54.0	50.5	46.6	50.4
Izera	–	8.91	8.95	8.93	–	48.6	43.9	46.3
Kandela	8.04	9.29	9.22	8.85	52.4	51.7	47.1	50.4
Katoda	8.46	9.10	8.68	8.74	53.6	48.7	47.1	49.8
KWS Torridon	–	9.87	8.77	9.32	–	45.3	46.4	45.9
Łagwa	8.12	8.62	9.22	8.65	53.8	49.7	50.3	51.3
Ostka Smolicka	8.80	8.62	8.41	8.61	54.4	47.6	47.3	49.8
Parabola	8.12	–	–	8.12	56.0	–	–	56.0
Radocha	8.12	8.14	–	8.13	55.2	51.7	–	53.5
SMH 87	6.09	–	–	6.09	53.2	–	–	53.2
Struna	–	–	9.04	9.04	–	–	45.5	45.5
Trappe	8.97	10.15	9.13	9.41	49.6	44.5	40.3	44.8
Tybalt	8.63	9.48	9.22	9.11	54.0	50.0	47.2	50.4
Waluta	7.87	8.43	8.50	8.26	51.2	48.5	46.5	48.7

Table 8. Resistance category of spring wheat cultivars determined from assessments, in 2011–2013

Cultivar	Cultivar response		FHB [%]	FDK [%]
	based on FHB incidence	based on FDK		
Arabella	moderately resistant	moderately susceptible	8.3	15.0
Bombona	moderately resistant	susceptible	9.2	21.7
Hewilla	moderately resistant	susceptible	7.3	24.3
Izera	moderately resistant	moderately susceptible	9.5	13.0
Kandela	moderately resistant	moderately susceptible	6.7	16.8
Katoda	moderately resistant	susceptible	10.5	30.7
KWS Torridon	moderately resistant	susceptible	11.8	23.0
Łagwa	moderately resistant	susceptible	5.3	22.3
Monsun	moderately resistant	moderately susceptible	11.0	20.0
Nawra	moderately resistant	susceptible	9.0	25.5
Ostka Smolicka	moderately resistant	moderately susceptible	5.7	19.3
Parabola	moderately resistant	susceptible	13.5	27.5
Radocha	moderately resistant	susceptible	11.5	26.0
SMH 87	moderately resistant	susceptible	13.5	23.0
Struna	moderately resistant	moderately susceptible	10.0	19.0
Trappe	moderately resistant	susceptible	11.8	28.0
Tybalt	moderately resistant	susceptible	16.3	35.3
Waluta	moderately resistant	susceptible	9.0	24.5

FHB – Fusarium head blight; FDK – *Fusarium*-colonised kernels

Arabella, Izera, Kandela, Monsun, Ostka Smolicka, and Struna expressed 'moderate susceptibility' and cvs Bombona, Hewilla, Katoda, KWS Torridon, Łagwa, Nawra, Parabola, Radocha, SMH 87, Trappe, Tybalt, and Waluta expressed 'susceptibility' when assessed by proportion of FDK. Cultivars differed in their levels of 'moderate resistance', 'moderate susceptibility', and 'susceptibility'.

Discussion

Growing wheat cultivars with greater resistance to FHB is the most promising strategy for disease control. Fusarium head blight resistance is, however, a complex trait. To date, sources of resistance conferring complete resistance to FHB have not been identified in wheat. Resistance to FHB has been shown to be under the control of a few major QTLs operating together with unknown numbers of minor genes (Buerstmayr *et al*. 2009; Kollers *et al*. 2013).

Reported sources of FHB resistance in spring wheat include a few landraces, 'Sumai 3' and its derivatives from China, 'Nobeoka Bozu' and 'Sin Chunaga' and its relatives from Japan, and 'Frontana' and 'Encruzilhada' from Brazil (Mesterhazy 1987; Liu and Wang 1991; Ban and Suenaga 2000; Yu *et al*. 2006).

Most success has been made in transferring FHB resistance from the Chinese wheat cultivar Sumai 3 (Anderson *et al*. 2001, 2007; Rudd *et al*. 2001). Cultivar Sumai 3 has the FHB resistance gene *Fhb1*, which has been extensively used in breeding programs as a major source of partial resistance to FHB. It showed a major effect on Type II resistance across different genetic backgrounds and environments. Other QTLs for FHB resistance have exhibited minor effects and their expression varied significantly.

Heavy use of narrow FHB resistance sources may increase selection pressure on the pathogens to erode the efficacy of the resistance genes involved. Thus, identification and characterisation of additional sources of resistance are important for enhancing the level of resistance and for introducing genetic variation to the breeding materials. Evaluation of FHB resistance in local cultivars of spring wheat may provide: (i) good lines for local breeding programmes and (ii) selected resistant cultivars for commercial farming aimed at production of mycotoxin-free grain in integrated FHB management systems based on forecasting models.

Spring wheat cultivars in Europe have the highest levels of resistance to FHB when compared with cultivars from South America or Asia (Zhang *et al*. 2008). Polish cultivars and foreign cultivars/lines have been continuously screened for FHB resistance (Góral 2005; Wiśniewska and Kowalczyk 2005; Lenc and Sadowski 2011; Góral and Walentyn-Góral 2014). Research has also concentrated on how the previous crop and weather conditions affect infection by *Fusarium*, and on concentrations of mycotoxins in grain (Sadowski *et al.* 2011; Góral *et al*. 2012). The most resistant genotypes, with acceptable agronomic characters, are used in farming and breeding, often despite the failure to identify their sources of resistance.

The results reported here showed significant differences in FHB incidence among spring wheat cultivars included in the Polish National List of Agricultural Plant Varieties, in natural non-epidemic conditions, in northern Poland, from 2011 to 2013. The official catalogues of varieties describe the FHB resistance of particular cultivars as average (cv. Łagwa), moderate (cvs Arabella, Hewilla, Kandela, Katoda, KWS Torridon, Monsun, Nawra, Ostka Smolicka, Parabola, Radocha, SMH 87, Trappe, Tybalt, and Waluta) or very good (cvs Bombona, Izera, and Struna). The average FHB severity, which in Lisewo ranged from 2.2–6.9%, is consistent with the general resistance to *Fusarium* spp., which would have contributed to the 2011–2013, non-epidemic situatioIt should be noted, that there were significant differences within this 'general resistance'. Only cv. Tybalt had FHB severity (DS) approaching 7%, which is the lower limit for light epidemics (Del Ponte *et al*. 2005). The cultivars compared were being recommended for commercial production in northern Poland in 2014 (Anonymous 2014). Cultivars Bombona, Hewilla, Kandela, Katoda, Łagwa, Monsun, Nawra Ostka Smolicka, Parabola, Trappe, and Waluta were also being used for the production of seeds. Each cultivar was on an area of 51–445 ha (Góral and Walentyn-Góral 2014). Choosing more susceptible cultivars would be associated with a greater risk of *Fusarium* mycotoxin contamination, a decreased germination rate, and poor seedling growth (Gilbert and Tekauz 1995). The use of infected grain can, by addition of virulent toxigenic biotypes of fungi, additionally enrich the complex of soil phytopathogens associated with seedling blight.

Fusarium head blight (FHB) is a major fungal disease in durum wheat. Fewer sources of resistance have been found in tetraploid durum wheat than in hexaploid wheat (Rudd *et al*. 2001; Oliver *et al.* 2008). There was high FHB incidence (13.5%) and disease severity (5.2%) in the single durum wheat cultivar (SMH 87) that we tested. The high FHB incidence was associated with the smallest grain yield (6.09 t · ha^{-1}). Durum wheat is said to be susceptible mostly to *F. graminearum* (*G. zeae*). It was suggested by Lionetti *et al.* (2015) that content and composition of cell wall polymers affect susceptibility to the wall-degrading enzymes produced by *F. graminearum* during infection, which affects the outcome of host-pathogen interactions. This fungus was not found in cv. SMH 87. Infected kernels were colonised mostly by *G. avenacea*.

Resistance of Type I (to initial infection), Type II (to spread within the head) and Type IV (to kernel infection) to FHB, were assessed in this study. Types I and II were assessed from FHB incidence and DS on developing heads, and Type IV from the proportion of FDK as suggested by Mesterhazy *et al.* (1999). Type III resistance (resistance to DON accumulation) is also said to be an important component of FHB resistance (Miller *et al*. 1985; Snijders and Perkowski 1990) but was not included in our study. Fusarium head blight incidence is not always correlated with DON concentration (Mesterházy *et al*. 1999; Bai *et al*. 2001; Koch *et al*. 2006; Brennan *et al*. 2007; Lehoczki-Krsjak *et al*. 2010; Wegulo *et al*. 2011; Gromadzka *et al.* 2012). It was assumed here, that cultivars with low kernel colonisation by *Fusarium* spp. had low concentrations of mycotoxins and stable FHB resistance (Snijders and Krechting 1992; Bai *et al*. 2001).

The assessments based on FHB incidence and DS confirmed general 'moderate FHB resistance' of the cultivars included in the study. The category 'moderately resistant' is based, however, on a wide range (5–25%) of FHB incidence values. On this basis, expression of resistance within the 'moderate resistance' category, was greatest in cvs Arabella, Bombona, Hewilla, Izera, Kandela, Łagwa, Nawra, Ostka Smolicka, Struna, and Waluta (FHB = 5.3–10.0%, DS = 2.2–5.0%). Less resistance in this 'moderate resistance' category was expressed by cvs Katoda, KWS Torridon, Monsun, Parabola, Radocha, SMH 87, Trappe, and Tybalt (FHB = 10.5–16.3%, DS = 3.6–6.9%). On the basis of the proportion of FDK, the cultivars were categorised as 'moderately susceptible/susceptible', i.e. with lower levels of resistance. These assessments of cultivar response to *Fusarium* infection in the field differ from the results of Zhang *et al*. (2008). They found that European cultivars of spring wheat with 'moderate resistance' in the field displayed higher levels of resistance based on the proportion of FDK.

There was a positive correlation between FHB incidence and the proportion of FDK. In more resistant cultivars, there was less kernel colonisation. An explanation is that limited fungal colonisation of kernels in resistant cultivars results from membrane-based tolerance of trichothecene mycotoxins (Snijders and Krechting 1992). However, a lack of significant correlation between FHB incidence and the proportion of FDK has also been reported, and attributed to two different sources of resistance (Sneller *et al*. 2012).

None of the cultivars in the present study was highly resistant to FHB. This is partly in agreement with Góral and Walentyn-Góral (2014) who found statistically significant variability in FHB resistance and a wide range of FHB incidences among 25 spring wheat cultivars from the Polish National List and 35 cultivars/lines from a collection of resistant forms over a three-year study (2010–2012). They found moderate FHB resistance in cvs Bombona, Łagwa, Monsun, Ostka Smolicka, Trappe, and Waluta, and greater resistance in cvs Hewilla, Kandela, Katoda, Nawra, and Parabola.

Fusarium poae, F. avenaceum, and *F. culmorum* were the most frequent *Fusarium* spp. in spring wheat kernels. They were also the most frequent in a study on winter wheat in Poland (Lenc *et al*. 2011; Lenc 2015). These species, together with *F. graminearum*, *F. pseudograminearum* O'Donnell & T. Aoki (teleomorph *Gibberella coronicola* T. Aoki & O'Donnell), *Microdochium nivale* (Fr.) Samuels & I.C. Hallett (teleomorph *Monographella nivalis* (Schaffnit) E. Müll.), and *M. majus* (Wollenw.) Glynn & S.G. Edwards, form the dominant group in FHB populations in Europe, USA, and Canada (Wilcoxon *et al*. 1988; Gale *et al*. 2007; Alvarez *et al*. 2010). *Fusarium avenaceum, F. culmorum*, and *F. poae* are more adapted to cooler/wet/humid regions (Doohan *et al*. 2003), although Xu *et al*. (2008) also associated *F. poae* with relatively drier and warmer conditions. According to Rohácik and Hudec (2005), a high incidence and density of *F. poae* in warmer places results from its adaptability to agro-environmental conditions during grain formation.

In general terms, *F. poae* and *F. avenaceum* are relatively weaker pathogens than *F. graminearum* and *F. culmorum* (Wong *et al*. 1992; Fernandez and Chen 2005; Xu *et al*. 2007). Only individual isolates of *F. poae* are strongly aggressive (comparable with *F. culmorum* and *F. graminearum*) (Brennan *et al*. 2007). It is receiving increased interest because of its toxigenic potential (it produces trichothecenes of types A and B, aurofusarin, beauvericin, butenolide, culmorin, cyclonerodiol, enniatins, fusarin, and moniliformin) and the human and animal mycotoxicoses it can cause (De Nijs *et al*. 1996a, b; Thrane *et al*. 2004; Chełkowski *et al*. 2007).

Fusarium graminearum was relatively rare in the spring wheat cultivars although its increasing contribution to FHB in Poland has been observed (Czaban *et al*. 2011; Lenc 2015). In general, *F. graminearum* favors warmer weather. The increasing significance of *F. graminearum* is related to global warming (higher temperatures in spring and summer) and increased amounts of maize residues and fungal inoculum associated with increased production of maize for grain (Wakuliński and Chełkowski 1993). *Fusarium langsethiae* Torp and Nirenberg, a species with increasing significance in Poland (Łukanowski *et al.* 2008; Łukanowski and Sadowski 2008), was not recorded.

The colonisation of kernels by other pathogenic or saprotrophic fungi may increase the likelihood of further deterioration and additional chemical contamination of the grain. The taxa recorded (Table 6) are often known to be secondary invaders, which can cause serious storage mold problems. *Alternaria*, *Aspergillus*, and *Penicillium* spp. are known mycotoxin producers (Steyn 1995). Their presence in grain constitutes a potential additional health risk. Storage of cereals under warm and humid conditions may further increase mycotoxin content even when field infections were only light to moderate. *Trichoderma viride*, which is antagonistic towards *Fusarium* spp. with a potential to reduce inoculum development (Inch and Gilbert 2007), occurred sporadically.

The studies were carried out in only one location, but the complexity of resistance mechanisms and significant environment effects on FHB development require screening in a range of environments (Fuentes *et al.* 2005). The location in northern Poland, however, is in the main target area for spring wheat production because of the weather conditions. In addition, the 'moderately resistant' categorisation of the cultivars chosen for study ensures that they should have some stability under different epidemic conditions. According to Mesterházy (1995), the stability of plant reaction is connected to the resistance level; the most resistant genotypes are the most stable, and the most susceptible ones tend to be unstable.

Weather is usually the main determinant of FHB incidence (Parry *et al*. 1995; De Wolf *et al*. 2003; Lemmens *et al*. 2004; Xu *et al*. 2008). Epidemics of FHB are associated with multiple inoculation episodes and with coincidental wet periods. Higher humidity favors the growth and sporulation of *Fusarium* fungi which are spread by wind and rain splashes. Lower humidity helps plants to dry and prevents infection and colonisation. In this study, though, a decreased average proportion of FDK was observed with increased rainfall in July. There was intense and prolonged rain in July 2011–2013. Rain intensity, duration, and frequency, as well as the size and velocity of falling drops, affect the splash dispersal of spores and success of infection (Shin *et al.* 2014). Intense rain may have a decreased effective dispersal by washing off newly-dispersed spores, which therefore, did not contribute to increased *Fusarium* infection (Penet *et al.* 2014). Wind will increase primary rain-dispersal distance in a downwind direction and decrease it upwind, although it is accepted that wind-dispersal distances are generally longer than rain dispersal alone, and that pure splash dispersal is mostly local (Yang *et al.* 1990; Sache 2000). The epidemiology of FHB caused by *F. poae* and the pathogenś infection biology are less understood then other major FHB patogens (Stenglein 2009).

Conclusions

Eighteen spring wheat cultivars were all found to be 'moderately resistant' to 'moderately susceptible' to FHB in non-epidemic situations in the field. There were differences within the wide range of resistance/susceptibility categories. The cultivars showing most resistance (Arabella, Izera, Kandela, Monsun, Ostka Smolicka, and Struna) can be recommended for breeding programmes and for commercial farming with the aim of producing mycotoxin-free grain in integrated FHB management systems.

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