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# Fusarium head blight (FHB) and Fusarium populations in grain of winter wheat grown in different cultivation systems

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**Abstract:** Fusarium head blight (FHB) incidence, and colonisation of grain by *Fusarium* species on winter wheat grown in organic, integrated, and conventional systems as well as in monoculture, were studied locally in Poland, from 2002 to 2010. Fusarium head blight incidence differed throughout the study years. It was found to occur the most where rainfall was highest and where rainfall was the most prolonged before, during, and after flowering of wheat. Fusarium head blight incidence was generally less on wheat grown organically than on wheat grown in other systems. In some years, FHB was noted more in monocultures than in other systems. *Fusarium poae* was the most common species of FHB populations in wheat kernels, followed by *F. avenaceum* and *F. tricinctum*. Other species which occurred more rarely or sporadically were: *F. culmorum, F. equiseti, F. graminearum, F. langsethiae, F. oxysporum,* and *F. sporotrichioides*. There were found to be significant effects of the cropping system on grain colonisation by *Fusarium* in some years. There was a positive correlation between FHB incidence and number of kernels colonised and damaged by *Fusarium,* in all four systems. Inferences were drawn concerning the effects of different procedures in different production systems and the possible value for controlling FHB.

Key words: conventional system, cultivar, cultivation, Fusarium, Fusarium head blight (FHB), integrated system, monoculture, organic, wheat

#### Introduction

Fusarium species are notable among wheat pathogens in causing pre- and post-emergence seedling blight, stembase (crown) rot or Fusarium head blight (FHB). Fusarium species often contribute to significant loss in production. Fusarium head blight is one of the most important cereal diseases. It has emerged as a major threat to wheat and barley crops around the world (Leonard and Bushnell 2003). Fusarium head blight contributes not only to loss of grain yield but also to the loss of grain quality because of the grains' contamination with mycotoxins.

Seventeen species of *Fusarium* are known to contribute to disease. These are species with different climatic requirements and genetic and environmental adaptations, which allow infection of different parts of wheat plants at different developmental stages (Parry *et al.* 1995; Stępień and Chełkowski 2010; Wiśniewska *et al.* 2014). *Fusarium graminearum* Schwabe *Gibberella zeae* (Schwein.) Petch (telemorph) is the predominant causal agent of FHB in most areas of the world.

Wheat is susceptible to infection at flowering (GS 50) through to early dough development stages. Spores must come into contact with the floret. The fungus can, however, also enter through wounds caused by hail, birds or insects. Symptoms of FHB usually include premature bleaching and dying of spikelets and shrunken, wrinkled, discolored, lightweight kernels. The colonised grain may

however stay symptomless. Primary infection followed by sporulation of *Fusarium* can lead to secondary infections. The secondary infections can be especially problematic in uneven wheat stands with late flowering tillers. Infection will continue as long as weather conditions are favorable and wheat plants are at susceptible growth stages.

Populations of FHB fungi over-winter on seed and crop residue. Grains infected with the *Fusarium* fungi may reduce emergence and reduce vigor of seedlings. The result is seedling blight. Some *Fusarium* species can also survive on roots of other crops, including pulses and oilseeds

Deoxynivalenol (DON) and zearalenone (ZEA) are the most common mycotoxins associated with FHB (Vesonder and Goliński 1989). They may be produced in high amounts, especially *F. graminearum* and *F. culmorum*. Recently *F. poae* ocurs often on wheat grains and produceds several toxins, including nivalenol (NIV).

Compared with other mycotoxins, DON is one of the least toxic but can still result in reduced feed consumption or feed refusal, especially when fed to non-ruminants. Toxins may also contaminate human food, with implications for human health. Established limits for mycotoxins in feed and food affect economics and international trade.

Certain weather conditions favour FHB, for example, extended periods of high moisture or relative humidity

(> 90%), frequent rainfall, moderately high temperatures (between 15 and 30°C), and occurrence of air currents. When these conditions are present before, during, and after flowering for at least 2–3 days, there may be a tendency towards inoculum production, dispersal and transport of spores, infection of florets and colonisation of developing grains (e.g. Lacey *et al.* 1999). During prolonged periods of high humidity and moisture, infection may occur at lower temperatures. The prevalence of disease also depends on the agronomic practices, effectiveness of fungicides used, and host resistance (Bottalico and Perrone 2002; Goliński *et al.* 2002; Lenc *et al.* 2009; Sadowski *et al.* 2010).

In Poland, FHB has been observed each year on approximately 70% of the wheat fields. But the disease incidence in some years has been low (< 1% of heads colonised; Wakuliński *et al.* 1991).

Intensification of agricultural production has been strongly driven by increased use of fertilisers, irrigation water, agricultural machinery, pesticides, and land. Such intensification raises public concern about sustainability in agricultural systems, the quality of human health, and the state of the environment. There is a need to develop technologies and practices that do not have adverse effects on the environment, are accessible to and effective for farmers, and lead to improvements in food production. These should encourage farmers to change from conventional farming to alternative production methods, including organic farming.

The objective of the study was to assess: (1) the local incidence of FHB in winter wheat grown in four different production systems: organic, integrated and conventional, and monoculture; (2) the extent of colonisation of wheat kernels by *Fusarium* species; (3) the populations of FHB *Fusarium* fungi in colonised kernels; (4) and the effects of the production system, weather conditions, and wheat cultivar on FHB incidence. Any resulting information on the effects of crop management practices (type of preceding crop, cover crop, tillage, fungicides) applied in different systems of wheat production, is expected to contribute to identifying optimal planting strategies for limiting disease. The studies were carried out for nine years (2002–2010) to assess the effects of weather conditions on FHB incidence and severity.

# **Materials and Methods**

#### Site description

Six winter wheat (*Triticum aestivum* L.) cultivars were grown in experimental fields at the Institute of Soil Science and Plant Cultivation, the State Research Institute, in Osiny, Poland (51°52′02″ N, 22°05′25″ E). Cultivars Elena and Juma were sown for harvest in 2002. Cultivars Sukces and Zyta were sown for harvest from 2003 to 2007, and cvs. Legenda and Rywalka from 2008 to 2010. The soil was uniformly sandy loam. The soil characteristics are given in table 1.

A thirteen-ha field was divided into blocks of 5, 4, 3, and 1 ha, under organic, integrated and conventional systems, and monoculture, respectively. Each block was

sub-divided into 1-ha whole plots and each whole plot sub-divided into four replicate sub-plots (2,500 m<sup>2</sup>) for sampling. This non-randomised block design, also used in other studies (Lenc *et al.* 2012), was necessitated by practical constraints. The number of whole plots was determined by the number of crops in the rotation sequence.

In the organic system, the crop sequence was: potato, spring barley or spring wheat, white clover + forage grasses, clover + forage grasses, winter wheat. No fertilisation and no fungicides were used. In the integrated system, which included an extended rotation, natural and artificial fertilisation, and fungicides, the crop sequence was: potato, spring barley or spring wheat, faba bean, winter wheat. In the conventional system, which had limited rotation, artificial fertilisers and fungicides, the crop sequence was: spring barley or spring wheat, oilseed rape, winter wheat. In monoculture only, winter wheat was grown each year from 2002 to 2010. Phased-in crop sequences ensured that one whole plot of winter wheat in each system was available for sampling in the harvest year period of 2002-2010. Details of crop management procedures are shown in table 2. The winter wheat seed was sown one week after autumn ploughing.

In the 2002–2010 time period, the average temperatures in June were 16.4–19.1°C and in July 18.5–22.5°C, with the highest in 2007 (June) and 2006 (July) (Table 3). Monthly rainfall in June was 19.2–95.8 mm and in July was 20.7–106.5 mm. The wettest June was in 2009, and the wettest July in 2005. The number of days with rainfall ranged from 5 to 19 in June, and from 3 to 17 in July.

In June 2005 (low temperature and rainfall), June–July 2006 (low rainfall), and June 2008 (low rainfall in June) the weather conditions were generally unfavourable for FHB development. In June–July 2002, 2003, 2004, 2007, and 2009 the weather conditions were more favourable.

## Collection of samples and disease assessment

Each year (2002–2010), 400 heads ( $4 \times 100$ ) of wheat from each cultivation system were collected at the late milk to the early dough development stage (GS 77-83; Zadoks et al. 1974) from randomly chosen plants taken across a diagonal transect in each of the four replicate sub-plots. Fusarium head blight incidence in wheat heads and disease severity were evaluated visually. The assessment was made on 400 heads. Fusarium head blight incidence was determined as the proportion (%) of heads with symptoms. Symptoms on individual heads were assessed using this scale: 0 – no symptoms; 1 – 5% of the head's surface with symptoms; 2 - 6-10% with symptoms; 3 - 11-30% with symptoms; 4 - 31-50% with symptoms; 5 – more than 50% with symptoms. Disease severity (*DS*) was evaluated using the Townsend-Heuberger's formula (Townsend and Heuberger 1943):

$$DS(\%) = \frac{\sum (nv)}{NV} \times 100$$

where: n – degree of infection on the 0–5 scale, v – number of heads per category, V – total number of heads assessed, N – highest degree of infection.



**Table 1.** Characteristics of soils at Osiny in 2002–2010

Soil characteristics		2002	2003	2004	2005	2006	2007	2008	2009	2010
			Organic							
pH in H <sub>2</sub> O		6.40	6.78	6.41	6.88	6.72	6.68	6.41	6.51	6.68
Humus content [%]		1.54	1.38	1.54	1.55	1.46	1.64	1.40	1.62	1.63
	spring	71	123	52	63	99	50	170	103	90
Extractable soil nitrogen $NO_3$ + $+ NH_4$ [kg N · ha <sup>-1</sup> ] <sup>a</sup>	autumn	51	78	78	67	89	64	132	90	51
Extractable soil phosphorus [mg·	kg-1]b	8.17	10.73	6.98	8.47	7.83	7.17	11.48	8.33	10.53
Extractable soil potassium [mg · k	g <sup>-1</sup> ] <sup>b</sup>	5.81	7.12	4.68	5.06	4.38	5.26	8.16	4.30	6.27
Extractable soil magnesium [mg ·	kg <sup>-1</sup> ] <sup>c</sup>	9.23	7.66	6.71	7.64	8.16	11.17	8.63	7.43	10.63
			Integrated	d system	,					
pH in H <sub>2</sub> O		6.48	6.56	6.56	6.66	6.63	6.35	6.26	6.77	6.32
Humus content [%]		1.31	1.07	1.28	1.27	1.46	1.32	1.27	1.29	1.45
Extractable soil nitrogen NO <sub>2</sub> +	spring	55	79	61	56	83	48	79	61	56
+ NH <sub>4</sub> [kg N · ha <sup>-1</sup> ] <sup>a</sup>	autumn	50	196	77	94	77	74	124	125	43
Extractable soil phosphorus [mg·	kg-1]b	9.97	15.25	11.25	13.07	13.90	15.73	11.07	17.60	12.90
Extractable soil potassium [mg · k	g <sup>-1</sup> ] <sup>b</sup>	11.47	9.29	9.02	12.56	12.48	11.20	9.90	11.20	12.13
Extractable soil magnesium [mg ·	kg <sup>-1</sup> ] <sup>c</sup>	6.56	5.56	5.44	8.55	7.03	6.93	6.63	11.33	8.33
		C	onventio	nal systen	n					
pH in H <sub>2</sub> O		6.87	6.98	6.17	6.90	6.90	5.91	7.02	7.05	6.80
Humus content [%]		1.01	1.35	1.53	1.02	1.34	1.51	1.09	1.41	1.49
Extractable soil nitrogen NO <sub>3</sub> +	spring	48	111	88	48	174	42	90	61	73
$+ NH_4 [kg N \cdot ha^{-1}]^a$	autumn	65	brak	108	71	136	149	102	104	67
Extractable soil phosphorus [mg ·	kg-1]b	21.3	19.45	10.48	16.80	19.36	11.17	21.60	21.50	13.27
Extractable soil potassium [mg · kg	g <sup>-1</sup> ] <sup>b</sup>	10.74	13.29	13.99	11.12	11.88	16.07	13.30	15.80	16.90
Extractable soil magnesium [mg ·	kg <sup>-1</sup> ] <sup>c</sup>	4.29	4.89	6.27	4.82	5.97	8.13	6.03	7.07	9.87
			Monoc	ulture						
pH in H <sub>2</sub> O		5.94	5.82	5.85	6.22	6.02	6.09	6.22	6.25	6.25
Humus content [%]		1.11	1.05	1.17	1.09	1.10	1.15	1.19	1.19	1.23
Extractable soil nitrogen NO <sub>3</sub> +	spring	60	137	220	96	130	45	125	58	56
$+ NH_4 [kg N \cdot ha^{-1}]^a$	autumn	153	195	210	240	168	152	135	96	67
Extractable soil phosphorus [mg·	kg <sup>-1</sup> ] <sup>b</sup>	8.28	10.75	8.25	10.07	8.78	8.92	9.48	9.87	10.57
Extractable soil potassium [mg · k	g <sup>-1</sup> ] <sup>b</sup>	11.12	11.36	9.08	12.88	8.89	9.23	10.32	12.03	12.30
Extractable soil magnesium [mg ·	kg <sup>-1</sup> ] <sup>c</sup>	7.20	7.53	6.39	7.79	7.20	8.33	9.54	9.37	10.93

<sup>&</sup>lt;sup>a</sup> analysed with the Kjeldahl method

 $<sup>^{\</sup>rm b}$  analysed with the Egner-Riehm method

<sup>&</sup>lt;sup>c</sup> analysed with the Schachtschabel method

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Treatment	Organic system	Integrated system	Conventional system	Monoculture
Preceding crops	potato spring barley – to 2004 spring wheat – from 2005 white clover ( <i>Trifolium repens</i> ) + forage grasses clover + forage grasses winter wheat	potato spring barley – to 2004 spring wheat – from 2005 faba bean ( <i>Vicia faba</i> L.) winter wheat	spring barley – to 2004 spring wheat – from 2005 oilseed rape ( <i>Brassica napus</i> ) winter wheat	winter wheat
Cover crop	in different years various combination of buckwheat (Fagopyrum esculentum) [20 kg·ha <sup>-1</sup> ] + narrow leaf lupin (Lupinus angustifolius) [50 kg·ha <sup>-1</sup> ] + white mustard (Sinapis alba) [25 kg·ha <sup>-1</sup> ] + common vetch (Vicia sativa) [50 kg·ha <sup>-1</sup> ] + faba bean (Vicia faba) [50 kg·ha <sup>-1</sup> ] applied after wheat harvest	in different years various combination of buckwheat (Fagopyrum esculentum) [20 kg · ha-¹] + narrow leaf lupin (Lupinus angustifolius) [50 kg · ha-¹] + white mustard (Sinapis alba) [25 kg · ha-¹] + common vetch (Vicia sativa) [50 kg · ha-¹] + faba bean (Vicia faba) [50 kg · ha-¹] applied after wheat harvest	1	ı
Tillage	autumn – first plough (12–14 cm deep), disc harrowing, shredding of the cover crop, post-harvest tillage (grubber), disc harrowing, winter plough (24–26 cm deep)	autumn – first plough (12–14 cm deep), disc harrowing, shredding of the cover crop, post-harvest tillage (grubber), disc harrowing, winter plough (24–26 cm deep)	autumn – post-harvest tillage (grubber), disc harrowing, winter plough (24–26 cm deep)	autumn – post-harvest tillage (grubber), disc harrowing, winter plough (24–26 cm deep)
Inorganic fertilizers	potassium sulphate, in autumn: 2002 – 80 kg K <sub>2</sub> O· ha <sup>-1</sup> 2004 – 50 kg K <sub>2</sub> O· ha <sup>-1</sup> 2005 – 60 kg K <sub>2</sub> O· ha <sup>-1</sup> 2006 – 50 kg K <sub>2</sub> O· ha <sup>-1</sup> 2007 – 75 kg K <sub>2</sub> O· ha <sup>-1</sup> 2008 – 75 kg K <sub>2</sub> O· ha <sup>-1</sup> 2009 – 75 kg K <sub>2</sub> O· ha <sup>-1</sup> 2010 – 75 kg K <sub>2</sub> O· ha <sup>-1</sup> 2010 – 75 kg K <sub>2</sub> O· ha <sup>-1</sup> 2010 – 75 kg K <sub>2</sub> O· ha <sup>-1</sup> 2010 – 75 kg K <sub>2</sub> O· ha <sup>-1</sup> 2010 – 75 kg P <sub>2</sub> O· ha <sup>-1</sup> 2010 – 75 kg P <sub>2</sub> O· ha <sup>-1</sup> 2010 – 27 kg PO <sub>4</sub> · ha <sup>-1</sup> 2010 – 27 kg PO <sub>4</sub> · ha <sup>-1</sup>	Politoska (NPK + Mg + S) or ammonium nitrate (NH <sub>4</sub> NO <sub>3</sub> + CaCO <sub>3</sub> + MgCO <sub>3</sub> ), in autumn  2002 – 105 kg NH <sub>4</sub> · ha <sup>-1</sup> , 50 kg P <sub>2</sub> O <sub>5</sub> · ha <sup>-1</sup> + 75 kg K <sub>2</sub> O · ha <sup>-1</sup> 2003 – 125 kg NH <sub>4</sub> · ha <sup>-1</sup> , 50 kg P <sub>2</sub> O <sub>5</sub> · ha <sup>-1</sup> + 75 kg K <sub>2</sub> O · ha <sup>-1</sup> 2004 – 80 kg NH <sub>4</sub> · ha <sup>-1</sup> , 50 kg P <sub>2</sub> O <sub>5</sub> · ha <sup>-1</sup> + 75 kg K <sub>2</sub> O · ha <sup>-1</sup> 2005 – 170 kg NH <sub>4</sub> · ha <sup>-1</sup> , 66 kg P <sub>2</sub> O <sub>5</sub> · ha <sup>-1</sup> + 99 kg K <sub>2</sub> O · ha <sup>-1</sup> 2006 – 164 kg NH <sub>4</sub> · ha <sup>-1</sup> , 66 kg P <sub>2</sub> O <sub>5</sub> · ha <sup>-1</sup> + 60 kg K <sub>2</sub> O · ha <sup>-1</sup> 2007 – 116 kg NH <sub>4</sub> · ha <sup>-1</sup> , 60 kg P <sub>2</sub> O <sub>5</sub> · ha <sup>-1</sup> + 80 kg K <sub>2</sub> O · ha <sup>-1</sup> 2009 – 98 kg NH <sub>4</sub> · ha <sup>-1</sup> , 60 kg P <sub>2</sub> O <sub>5</sub> · ha <sup>-1</sup> 2009 – 98 kg NH <sub>4</sub> · ha <sup>-1</sup> , 60 kg P <sub>2</sub> O <sub>5</sub> · ha <sup>-1</sup> + 78 kg K <sub>2</sub> O · ha <sup>-1</sup> 2010 – 113 kg NH <sub>4</sub> · ha <sup>-1</sup> , 60 kg P <sub>2</sub> O <sub>5</sub> · ha <sup>-1</sup> + 90 kg K <sub>2</sub> O · ha <sup>-1</sup>	Foliroska (NPK + Mg + S) or ammonium nitrate (NH <sub>4</sub> NO <sub>3</sub> + CaCO <sub>3</sub> + MgCO <sub>3</sub> ), in autumn  2002 – 149 kg NH <sub>4</sub> · ha <sup>-1</sup> , 60 kg P <sub>2</sub> O <sub>5</sub> · ha <sup>-1</sup> + 90 kg K <sub>2</sub> O · ha <sup>-1</sup> 2003 – 168 kg NH <sub>4</sub> · ha <sup>-1</sup> , 60 kg P <sub>2</sub> O <sub>5</sub> · ha <sup>-1</sup> + 90 kg K <sub>2</sub> O · ha <sup>-1</sup> 2004 – 164 kg NH <sub>4</sub> · ha <sup>-1</sup> , 72 kg P <sub>2</sub> O <sub>5</sub> · ha <sup>-1</sup> + 72 kg K <sub>2</sub> O · ha <sup>-1</sup> 2005 – 105 kg NH <sub>4</sub> · ha <sup>-1</sup> , 72 kg P <sub>2</sub> O <sub>5</sub> · ha <sup>-1</sup> + 75 kg K <sub>2</sub> O · ha <sup>-1</sup> 2006 – 164 kg NH <sub>4</sub> · ha <sup>-1</sup> , 72 kg P <sub>2</sub> O <sub>5</sub> · ha <sup>-1</sup> + 75 kg K <sub>2</sub> O · ha <sup>-1</sup> 2007 – 173 kg NH <sub>4</sub> · ha <sup>-1</sup> , 70 kg P <sub>2</sub> O <sub>5</sub> · ha <sup>-1</sup> + 96 kg K <sub>2</sub> O · ha <sup>-1</sup> 2009 – 140 kg NH <sub>4</sub> · ha <sup>-1</sup> , 70 kg P <sub>2</sub> O <sub>5</sub> · ha <sup>-1</sup> + 98 kg K <sub>2</sub> O · ha <sup>-1</sup> 2010 – 151 kg NH <sub>4</sub> · ha <sup>-1</sup> , 70 kg P <sub>2</sub> O <sub>5</sub> · ha <sup>-1</sup> + 105 kg K <sub>2</sub> O · ha <sup>-1</sup>	Politoska (NPK + Mg + 5) or ammonium nitrate (NH <sub>4</sub> NO <sub>3</sub> + CaCO <sub>3</sub> + MgCO <sub>3</sub> ), in autumn  2002 – 219 kg NH <sub>4</sub> · ha <sup>-1</sup> , 60 kg P <sub>2</sub> O <sub>5</sub> · ha <sup>-1</sup> + 90 kg K <sub>2</sub> O · ha <sup>-1</sup> 2003 – 215 kg NH <sub>4</sub> · ha <sup>-1</sup> , 72 kg P <sub>2</sub> O <sub>5</sub> · ha <sup>-1</sup> + 72 kg K <sub>2</sub> O · ha <sup>-1</sup> 2004 – 154 kg NH <sub>4</sub> · ha <sup>-1</sup> , 72 kg P <sub>2</sub> O <sub>5</sub> · ha <sup>-1</sup> + 72 kg K <sub>2</sub> O · ha <sup>-1</sup> 2005 – 194 kg NH <sub>4</sub> · ha <sup>-1</sup> , 72 kg P <sub>2</sub> O <sub>5</sub> · ha <sup>-1</sup> + 72 kg K <sub>2</sub> O · ha <sup>-1</sup> 2006 – 199 kg NH <sub>4</sub> · ha <sup>-1</sup> , 72 kg P <sub>2</sub> O <sub>5</sub> · ha <sup>-1</sup> + 99 kg K <sub>2</sub> O · ha <sup>-1</sup> 2007 – 157 kg NH <sub>4</sub> · ha <sup>-1</sup> , 72 kg P <sub>2</sub> O <sub>5</sub> · ha <sup>-1</sup> + 75 kg K <sub>2</sub> O · ha <sup>-1</sup> 2007 – 157 kg NH <sub>4</sub> · ha <sup>-1</sup> , 70 kg P <sub>2</sub> O <sub>5</sub> · ha <sup>-1</sup> + 75 kg K <sub>2</sub> O · ha <sup>-1</sup> 2009 – 160 kg NH <sub>4</sub> · ha <sup>-1</sup> , 70 kg P <sub>2</sub> O <sub>5</sub> · ha <sup>-1</sup> + 98 kg K <sub>2</sub> O · ha <sup>-1</sup> 2010 – 136 kg NH <sub>4</sub> · ha <sup>-1</sup> , 70 kg P <sub>2</sub> O <sub>5</sub> · ha <sup>-1</sup> + 1165 kg K 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			18 April 1919 1919 1919 1919 1919 1919 1919 19	<u>ə</u>	) on 10 iroxamine fune 30 SC) on	VG) on 12 pendazim <sup>1</sup> j and nol + on 8 June	VG) on ) on 2 June , flusilasole June
Monoculture	175 kg NH <sub>4</sub> · ha <sup>-1</sup> , 69 kg P <sub>2</sub> O <sub>5</sub> · ha <sup>-1</sup> + 87 kg K <sub>2</sub> O · ha <sup>-1</sup>		2002 – benomyl (Benlate 50 WP) on 18 April [0.3 kg · ha <sup>-1</sup> ], triadimenol + tebuconazole (Folicur Plus 375 EC) on 16 May [1 1 · ha <sup>-1</sup> ], tebuconazole + spiroxamine + triadimenol (Falcon 460 EC) on 3 June [1 1 · ha <sup>-1</sup> ]	2003 – carbendazim (Sarfun 500 SC) + triadimefon (Bayleton Total 37.5 WP) on 26 May [1 kg · ha <sup>-</sup> l], triadimenol + tebuconazole (Folicur Plus 375 EC) on 6 June [1 l · ha <sup>-</sup> l], tebuconazole + spiroxamine + triadimenol (Falcon 460 EC) on 16 June [1 l · ha <sup>-</sup> l]	2004 – carbendazim (Sarfun 500 SC) on 10 May [0.51 · ha <sup>-1</sup> ], tebuconazole + spiroxamine + triadimenol (Falcon 460 EC) on 7 June [11 · ha <sup>-1</sup> ], picoxystrobin (Akanto 250 SC) on 4 June [0.51 · ha <sup>-1</sup> ]	2005 – carbendazim (Bavistin 500 WG) on 12 May [0.4 kg·ha-l], flusilasole + carbendazim (Alert 375 SC) on 12 May [0.61·ha-l] and on 25 May 2005 [11·ha-l], triadimenol + tebuconazole (Folicur Plus 375 EC) on 8 June [0.81·ha-l]	2006 – carbendazim (Bavistin 500 WG) on 16 May [0.3 kg · ha-¹] (Artea 330 EC) on 2 June [0.51 · ha-¹], flusilasole + carbendazim (Alert 375 SC) on 23 June [11 · ha-¹]
Conventional system	152 kg NH <sub>4</sub> · ha <sup>-1</sup> , 66 kg P <sub>2</sub> O <sub>5</sub> · ha <sup>-1</sup> + 86 kg 175 K <sub>2</sub> O · ha <sup>-1</sup> $K_2$ O · ha <sup>-1</sup>		2002 – carbendazim (Sarfun 500 SC) + 2002 triadimefon (Bayleton Total 37.5 WP) on 30 [0.3 April [1 kg · ha <sup>-1</sup> ], triadimenol + tebuconazole triad (Folicur Plus 375 EC) on 21 May [11 · ha <sup>-1</sup> ], 375 tebuconazole + spiroxamine + triadimenol + spiroxamine (Falcon 460 EC) on 3 June [11 · ha <sup>-1</sup> ] on 3	2003 – triadimenol + tebuconazole (Folicur Plus 375 EC) on 6 June [11 · ha <sup>-</sup> l], tebuconazole + spiroxamine + triadimenol (Falcon 460 EC) on 16 June [11 · ha <sup>-</sup> l]	2004 – carbendazim (Sarfun 500 SC) on 2000 10 May [0.51 · ha-l], propiconazole + cyproconazole (Artea 330 EC) on 19 May [0.41 + tri · ha-l], propiconazole + fenpropidin (Tilt Plus [11. + 400 EC) on 19 May [0.61 · ha-l], tebuconazole + 4 Ju spiroxamine + triadimenol (Falcon 460 EC) on 7 June [11 · ha-l]	2005 – carbendazim (Bavistin 500 WG) on 12 2008  May [0.4 kg · ha <sup>-1</sup> ], flusilasole + carbendazim May  (Alert 375 SC) on 12 May [0.61 · ha <sup>-1</sup> ],  triadimenol + tebuconazole (Folicur Plus 375 on 2  EC) on 9 June [0.81 · ha <sup>-1</sup> ]  [0.8]	2006 – carbendazim (Bavistin 500 WG) on 16 May [0.3 1 · ha <sup>-1</sup> ], propiconazole + cyproconazole (Artea 330 EC) on 16 May [0.5 1 · ha <sup>-1</sup> ] and 2 June [0.51 · ha <sup>-1</sup> ], flusilasole + ca + carbendazim (Alert 375 SC) on 23 June [11]
Integrated system	$120 \text{ kg NH}_4 \cdot \text{ha}^{-1}$ , 57 kg $\text{P}_2\text{O}_5 \cdot \text{ha}^{-1}$ + 77 kg $\text{K}_2\text{O} \cdot \text{ha}^{-1}$	acompost – under potato, in October – [30 dt·ha-1]	2002 – carbendazim (Sarfun 500 SC) + triadimefon (Bayleton Total 37.5 WP) on 30 April [0.8 kg · ha-¹], tebuconazole + spiroxamine + triadimenol (Falcon 460 EC) on 4 May [11 · ha-¹]	2003 – triadimenol + tebuconazole (Folicur Plus 375 EC) on 6 June [11 · ha <sup>-1</sup> ], tebuconazole + spiroxamine + triadimenol (Falcon 460 EC) on 16 June [11 · ha <sup>-1</sup> ]	2004 – triadimenol + tebuconazole (Folicur Plus 375 EC) on 4 June [0.351·ha <sup>-1</sup> ], picoxystrobin (Akanto 250 SC) on 4 June [0.351·ha <sup>-1</sup> ]	2005 – carbendazim (Bavistin 500 WG) on 12 May [0.2 kg · ha-1], flusilasole + carbendazim (Alert 375 SC) on 12 May [0.3 I · ha-1], 23 June [0.8 I · ha-1], triadimenol + tebuconazole (Folicur Plus 375 EC) on 8 June [0.4 I · ha-1], propiconazole + fenpropidin (Tilt Plus 400 EC) on 23 June [0.4 I · ha-1]	2006 – carbendazim (Bavistin 500 WG) on 16 May [0.4 kg · ha <sup>-1</sup> ], propiconazole + cyproconazole (Artea 330 EC) on 28 June [0.4 l · ha <sup>-1</sup> ]
Organic system	69 kg $K_2O \cdot ha^{-1}$ 11 kg $PO_4 \cdot ha^{-1}$	*compost – under potato, in October – [30 dt · ha-1]					
Treatment	Inorganic fertilizers (on average)	Organic fertilizers	Fungicides				

Table 2. Crop management procedures used in different systems of wheat production at Osiny in 2002-2010 - continuation

Treatment	Organic system	Integrated system	Conventional system	Monoculture
		2007 – spiroxamine (Impuls 500 EC) on 12 April [11 · ha <sup>-</sup> ], carbendazim (Karben 500 SC) on 12 April [0.51 · ha <sup>-</sup> ], protioconazole + fluoxastrobin (Fandango 200 EC), on 20 May [11 · ha <sup>-</sup> ]	2007 – spiroxamine (Impuls 500 EC) on 12 April [11 ha <sup>-1</sup> ], carbendazim (Karben 500 SC) on 12 April [0.51 ha <sup>-1</sup> ], protioconazole + fluoxastrobin (Fandango 200 EC), on 20 May [11 ha <sup>-1</sup> ]	2007 – spiroxamine (Impuls 500 EC) on 12 April [11 · ha <sup>-1</sup> ], carbendazim (Karben 500 SC) on 12 April [0.51 · ha <sup>-1</sup> ], famoxat + flusilazol (Charisma 207 EC) on 25 May [1.51 · ha <sup>-1</sup> ], protioconazole + fluoxastrobin (Fandango 200 EC), on 10 June [11 · ha <sup>-1</sup> ]
		2008 – propiconazole + fenpropidin (Tilt Plus 400 EC) on 7 May [1 1 · ha <sup>-</sup> 1], cyprodinil (Unix 75 WG) on 7 May [0.7 1 · ha <sup>-</sup> 1], protioconazol + tebuconazol (Prosaro 250 EC) on 30 May [1 1 · ha <sup>-</sup> 1]	2008 – propiconazole + fenpropidin (Tilt Plus 400 EC) on 7 May [11 · ha <sup>-1</sup> ], cyprodinil (Unix 75 WG) on 7 May [0.7 1 · ha <sup>-1</sup> ], tebuconazol (Prosaro 250 EC) on 30 May [1 1 · ha <sup>-1</sup> ]	2008 – propiconazole + fenpropidin (Tilt Plus 400 EC) on 7 May [11·ha-1], cyprodinil (Unix 75 WG) on 7 May [0.71·ha-1], tebuconazol (Prosaro 250 EC) on 30 May [11·ha-1]
		2009 – prolioconazol + spiroxamine (Input 460 EC) on 7 May [0.8 l·ha <sup>-1</sup> ], protioconazol + tebuconazol (Prosaro 250 EC) on 2 June [0.8 l·ha <sup>-1</sup> ]	2009 – prolioconazol + spiroxamine (Input 460 EC) on 7 May [1 1· ha <sup>-</sup> 1], protioconazol + tebuconazol (Prosaro 250 EC) on 2 June [1 1· ha <sup>-</sup> 1]	2009 – prolioconazol + spiroxamine (Input 460 EC) on 7 May [1 I·ha <sup>-1</sup> ], protioconazol + tebuconazol (Prosaro 250 EC) on 2 June [1 I·ha <sup>-1</sup> ]
		400 EC) on 29 April [0.91 · ha-1], thiophanate-methyl + tetraconazole (Yamato 303 SE) on 7 June [2 1 · ha-1]	2010 – fenpropimorph (Corbel 750 EC) on 7 April [11 · ha <sup>-</sup> ], propiconazole + fenpropidin (Tilt Plus 400 EC) on 30 April [0.9 I · ha <sup>-</sup> ], thiophanate-methyl + tetraconazole (Yamato 303 SE) on 7 June [2 I · ha <sup>-</sup> ]	2010 – fenpropimorph (Corbel 750 EC) on 7 April [11 · ha-1], propiconazole + fenpropidin (Tilt Plus 400 EC) on 30 April [0.91 · ha-1], thiophanate-methyl + tetraconazole (Yamato 303 SE) on 7 June [21 · ha-1]
Removal of weeds	harrowing or manually during vegetation	harrowing	harrowing	harrowing

a compost included solid cattle manure enriched with grasses and clover and provided 2.8 dt · ha-1 of organic matter and 0.1 dt · ha-1 of N, 0.04 dt · ha-1 of P, 0.1 dt · ha-1 of K, 0.25 dt · ha-1 of Ca, 0.27 dt · ha-1 of Mg, 0.005 dt  $\cdot$  ha<sup>-1</sup> of Na



Table 3. Temperature and rainfall during flowering and ripening stages of winter wheat growth at Osiny in 2002–2010

Mounth	Decade	2002	2003	2004	2005	2006	2007	2008	2009	2010	1871–1996
				Mean	temperatu	re [°C]	-				
	I	16.1	20.1	16.5	13.7	12.3	18.9	18.8	14.7	19.4	
June	II	19.1	17.7	16.3	17.3	18.5	20.8	16.9	15.4	18.2	16.7
June	III	18.8	16.9	16.6	18.2	22.2	17.7	19.0	19.6	17.4	16.7
	I–III	18.0	18.2	16.5	16.4	17.7	19.1	18.2	16.6	18.3	
	I	21.8	18.0	17.6	19.9	22.3	17.1	18.2	20.2	20.7	
Teeler	II	22.7	19.9	17.7	20.6	21.5	21.2	18.7	20.6	24.9	18.4
July	III	20.3	22.9	20.0	20.1	23.6	19.6	19.4	19.5	20.7	10.4
	I–III	21.6	20.4	18.5	20.2	22.5	19.3	18.8	20.1	22.1	
				Tota	l rainfall [1	nm]					
	I	39.8	1.6	13.0	18.7	18.7	42.8	0.0	31.2	21.6	
June	II	36.4	27.6	31.8	12.9	0.0	6.4	26.1	27.4	16.4	70.0
june	III	12.2	17.2	7.3	0.1	0.5	13.6	16.2	37.2	9.8	70.0
	I–III	88.4	46.4	52.1	31.7	19.2	62.8	42.3	95.8	47.8	
	I	14.6	25.9	9.7	0.0	0.0	27.9	16.1	33.0	0.1	
Teeler	II	62.8	10.4	47.7	40.7	16.7	17.6	21.8	3.9	2.3	83.0
July	III	1.4	17.9	35.6	65.8	4.0	3.5	55.7	32.1	40.2	65.0
	I–III	78.8	54.2	93.0	106.5	20.7	49.0	93.6	69.0	42.6	
				Number (	of days wi	th rainfall					
	I	5	1	4	6	7	4	0	6	3	
Tumo	II	3	6	6	4	0	2	4	7	3	
June	III	5	8	6	1	1	5	1	6	2	_
	I–III	13	15	16	11	8	11	5	19	8	
	I	2	4	5	0	0	6	4	7	1	
Teeler	II	5	4	5	4	2	3	7	4	2	
July	III	1	3	6	8	1	2	4	6	6	_
	I–III	8	11	16	12	3	11	15	17	9	

## Colonisation of wheat kernels by fungi

Mycological analysis of four samples of 400 (4 × 100) wheat kernels collected randomly from each of the four systems were performed in each of the study years, from 2005 to 2010. Grains were collected at immediately after harvest. 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. The 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 representatives of the fungi were identified by morphotyping on potato dextrose agar (PDA) and synthetic nutrient agar (SNA; KH<sub>2</sub>PO<sub>4</sub> 1 g · l<sup>-1</sup>, KNO<sub>3</sub> 1 g · l<sup>-1</sup>, MgSO<sub>4</sub> · 7H<sub>2</sub>O 0.5 g · l<sup>-1</sup>, KCl  $0.5 \text{ g} \cdot l^{-1}$ , glucose  $0.2 \text{ g} \cdot l^{-1}$ , sucrose  $0.2 \text{ g} \cdot l^{-1}$ ) using Booth (1971) and Kwaśna et al. (1991).

The amount of colonisation was calculated as the proportion (%) of kernels colonised and damaged by *Fusarium* spp. in a 400-kernel sample.

## Statistical analysis

The statistical significance of differences in FHB incidence and in disease severity were tested by two-way analysis of variance (ANOVA,  $p \le 0.05$ ) and Tukey's post hoc test, with cultivation system and cultivar as the two variables, using Statgraphics<sup>TM</sup> Centurion (Statpoint Technologies Inc., Warrenton, VA, USA). Percentage values were transformed into Bliss degrees before statistical analysis. The statistical significance of difference between numbers of kernels colonised by Fusarium spp. from two different samples was determined by  $\chi^2$ -test. The null hypothesis assumed that wheat from different systems has the same number of kernels colonised by Fusarium spp. Pearson's correlation coefficient was applied to analyse the relationship between FHB incidence and temperature or rainfall, and between FHB incidence and colonisation of kernels by Fusarium spp. In the last analysis the number of diseased heads (FHB) and the number of Fusarium-colonised and damaged kernels (FDK) were the independent and dependent variables (X and Y, respectively). The following coefficients, shown with their formulae, were calculated:

– linear regression coefficient *b*:

$$b = \frac{\sum xy}{\sum x^2},$$

- coefficient a:

$$a = \frac{\sum Y - b \sum X}{N},$$

– correlation coefficient *r*:

$$r = \frac{\sum xy}{\sqrt{(\sum x^2)(\sum y^2)}},$$

– linear regression equation  $Y_n$ :

$$Y_p = a + bX$$
.

where: x, y – standard deviation from the average X and Y.

#### Results

#### Fusarium head blight assessment

Differences in FHB incidence and DS occurred locally (between cropping systems) and seasonally (from between 2002 to 2010); FHB incidence reached a maximum of 45% and DS 9% (Table 4). Disease occurred the most in 2002 (mean FHB incidence = 30.7% and mean DS = 5.9%), was moderate in 2004, 2007, and 2010 (mean FHB incidence = 8.0–15.5% and mean DS = 1.7–3.4%), and was lowest in 2003, and 2009 (mean FHB incidence = 1.8 and 3.1% and mean DS = 0.3 and 0.7%). In 2005, 2006, and 2008, FHB symptoms were absent, slight or only sporadic.

## Effects of the cultivation system on disease

In 2002, the integrated system and monoculture had the most disease. In these two systems, the mean FHB incidences were similar (36.0 and 38.3%, respectively) (Table 4). Disease severity in the integrated system (6.8%) was significantly less, however, than that in the monoculture (7.6%). Disease was significantly less in the organic and conventional systems (FHB = 27.2 and 21.3%; DS = 5.1and 4.2%). In 2003, the mean FHB incidence and DS were generally low, with the highest values in the monoculture (FHB = 3.5%, DS = 0.7%). In the organic, integrated, and conventional systems, FHB incidence and DS were similar (FHB = 0.5–1.8%, DS = 0.1–0.3%) and significantly less than in the monoculture. In 2004, the mean FHB incidence and DS were again significantly higher in the monoculture (FHB = 20.1%, DS = 3.6%) than in the organic, integrated, and conventional systems (FHB = 7.2-15.4%, DS = 1.3-2.7). In 2007, the mean FHB incidence and DS were moderately high. Values were similar in the conventional system and the monoculture (FHB = 20.0 and 20.3%, DS = 4.8 and 4.1%), and significantly higher than in the integrated and organic systems, the latter having the least amount of disease. In 2009, the mean FHB incidence and DS were less (FHB = 3.1%, DS = 0.7%) but

there was significantly more disease in the organic system (FHB = 5.0%, DS = 1.2%) than in the other systems, which had similar values (FHB = 2.4-2.6%, DS = 0.6%). In 2010, there was slightly more disease, which was similar in all systems (mean FHB = 8.0%, DS = 1.7%).

Averaged over all the study years (2002–2010), there was no significant effect of cultivation system on the mean FHB incidence or DS. The most disease (not significantly) was found in the monoculture (FHB = 10.4%, DS = 2.0%) and the least was found in the organic system (FHB = 6.4%, DS = 1.2%).

#### Effects of cultivar on disease

Differences between cultivars usually occurred when there was sufficient disease, but the differences were not consistent between years and cultivation systems (Table 4). There were significant differences between cultivars in FHB incidence, in 2002, 2004, 2007, and 2009. In 2002, mean FHB incidence was less on cv. Elena than on cv. Juma. This difference occurred in the integrated system and monoculture. Elena had more FHB in the conventional system; infection of cultivars was similar in the organic system. Cultivars Sukces and Zyta had similar amounts of disease when averaged over all the years (2003-2007). In 2004, however, there was significantly less disease on cv. Sukces than on Zyta in all systems. In 2007, there was significantly less disease on cv. Sukces only in the conventional system. Zyta had less disease in the organic system and monoculture. Cultivars Legenda and Rywalka differed significantly in the amounts of disease only in 2009, when less FHB occurred on cv. Legenda in the conventional system and the monoculture and, not significantly, in the integrated sytem, and on cv. Rywalka in the organic system.

## Effects of weather conditions on disease

Fusarium head blight incidence and DS were not correlated with temperature and only lowly correlated with rainfall in June (r = 0.249, r = 0.204-0.595,  $p \le 0.001$ , respectively). The higher FHB incidence in 2002, 2004, and 2007 was associated with more rainfall over an extended period in June and the first decade of July (during inflorescence emergence, flowering, and ripening of winter wheat) (Table 3). In 2007, the high FHB incidence occurred even with low total rainfall in July 2007 (only 60% of the long-term average) and a dry end of July (last 10 days). The very low FHB incidence in 2003, was associated with high temperatures and low rainfall. No FHB or only a sporadic incidence in 2005, 2006, and 2008, was associated with a relatively cool June but low rainfall in June-July. The low incidence of FHB in 2009, was associated with a relatively cool June but extended rainfall. Moderate FHB incidence in 2010, was associated with a very warm June but moderately dry conditions.

## Colonisation of kernels by Fusarium spp.

There were significant differences between cultivation systems in percentage of wheat kernels colonised and damaged by *Fusarium* sp. (FDK) in each of the study years from



Table 4. Fusarium head blight incidence (FHB) and disease severity (DS) on different cultivars of winter wheat at Osiny in 2002–2010

			FHB [%]			_		DS [%]		
Cultivar -	О	I	С	M	mean	0	I	С	M	mean
					2002	_				
Elena	28.8 ab	31.5* a	25.0* b	31.5* a	29.2*	5.2 b	6.3* a	5.2* b	6.2* ab	5.7
Juma	25.5 b	40.4* a	17.5* c	45.0* a	32.1*	4.9 c	7.3* b	3.2* d	9.0* a	6.1
Mean	27.2 b	36.0 a	21.3 c	38.3 a	30.7	5.1 c	6.8 b	4.2 d	7.6 a	5.9
					2003					
Sukces	1.0	1.0	0.5	3.0	1.4	0.2	0.2	0.1	0.5	0.3
Zyta	2.5	1.5	0.5	4.0	2.1	0.4	0.2	0.1	0.8	0.4
Mean	1.8 ab	1.3 b	0.5 b	3.5 a	1.8	0.3 b	0.2 b	0.1 b	0.7 a	0.3
					2004					
Sukces	0.5* d	12.3* b	6.5* c	17.8* a	9.3*	0.1* d	2.2* b	1.1* c	3.2* a	1.7*
Zyta	13.8*bc	18.5* ab	11.5* c	22.3* a	16.5*	2.4* c	3.2* b	2.0* c	4.0* a	2.9*
Mean	7.2 d	15.4 b	9.0 c	20.1 a	12.9	1.3 c	2.7 b	1.6 c	3.6 a	2.3
					2005					
Sukces	0.5	0.2	1.0	1.0	0.7	-	-	-	_	
Zyta	0.8	1.5	1.2	1.0	1.1	_	_	_	_	_
Mean	0.7	0.9	1.1	1.0	0.9	_	_	_	-	_
					2006					
Sukces			No	vricible crm	ntoms or o	nly sporadic	EUR incider	200		
Zyta			INC	visible sylli	ptoms or o	iny sporadic	riib incidei	ice		
					2007					
Sukces	9.5* c	15.0 bc	16.0* b	23.5* a	16.0	2.2* b	3.6 a	3.5* a	4.7* a	3.5
Zyta	4.5* c	14.5 b	24.0* a	17.0* ab	15.0	1.0* c	2.9 b	6.0* a	3.4* b	3.3
Mean	7.0 c	14.8 b	20.0 a	20.3 a	15.5	1.6 c	3.3 b	4.8 a	4.1 ab	3.4
				Mea	n in 2003–2	2007				
Sukces	2.3 c	5.7 ab	4.8 bc	9.1 a	5.5	0.5	1.2	0.9	1.7	1.1
Zyta	4.3 b	7.2 ab	7.4 ab	8.9 a	6.9	0.8	1.3	1.6	1.6	1.3
					2008					
Legenda	0.8	0.5	0.8	0.5	0.6	-	_	-	_	_
Rywalka	0.5	0.5	0.2	0.5	0.4	_	_	-	-	-
Mean	0.6	0.5	0.5	0.5	0.5	-	_	_	-	_
					2009					
Legenda	5.6* a	1.6 b	0.9* b	1.3* b	2.4*	1.4* a	0.4 b	0.2* b	0.3* b	0.6*
Rywalka	4.3*	3.3	4.2*	3.6*	3.9*	0.9*	0.8	0.9*	0.8*	0.9*
Mean	5.0 a	2.4 b	2.6 b	2.4 b	3.1	1.2 a	0.6 b	0.6 b	0.6 b	0.7
					2010					
Legenda	9.6	8.1	8.3	7.5	8.4	1.9	1.9	1.7	1.5	1.8
Rywalka	7.0	8.3	7.6	8.1	7.8	1.4	1.9	1.5	1.6	1.6
Mean	8.3	8.2	8.0	7.8	8.0	1.6	1.9	1.6	1.6	1.7
				Mea	n in 2008–2	2010				
Legenda	5.3	3.4	3.3	3.1	3.8	1.1	0.8	0.6	0.6	0.8
Rywalka	3.9	4.0	4.0	4.1	4.0	0.8	0.9	0.8	0.8	0.8
				Mea	n in 2002–2	2010				
All cultivars	6.4	8.8	7.0	10.4	8.2	1.2	1.7	1.4	2.0	1.6

O – organic, I – integrated, C – conventional, M – monoculture; different letter in a row or asterisk in a column, in individual year, indicates significant difference according to two-way ANOVA at  $p \le 0.05$ 

Least significant difference (LSD), at p = 0.05

 $2002-system = 0.75, cultivar = ns \ (not \ significant), \ cultivar/system = 0.79, \ system/cultivar = 1.06$ 

2003 – system = 0.36, cultivar = ns, cultivar/system = ns, system/cultivar = ns

 $2004-system=0.51, cultivar=0.27, \ cultivar/system=0.54, \ system/cultivar=0.72$ 

 $2007-system=0.91, cultivar=ns, \ cultivar/system=0.96, \ system/cultivar=1.29$ 

2009 - system = 0.45, cultivar = 0.24, cultivar/system = 0.48, system/cultivar = 0.64

2010 – system = ns, cultivar = ns, cultivar/system = ns, system/cultivar = ns

Table 5. Mean proportion (%) of kernels colonized by Fusarium spp. in different cultivation systems at Osiny in 2005–2010

System	2005	2006	2007	2008	2009	2010	2005–2010
Organic	4.0 b	3.0 b	15.1 b	1.3 bc	13.0 b	14.8 a	8.5 a
Integrated	5.0 a	4.7 b	26.0 a	2.5 b	14.5 b	3.3 b	9.3 a
Conventional	7.5 a	11.4 a	25.7 a	7.5 a	21.0 a	3.6 b	12.8 a
Monoculture	7.3 a	4.9 b	27.6 a	0.7 c	14.0 b	5.5 b	10.0 a

A different letter in a column indicates significant difference according to  $\chi^2$  -test at  $p \leq 0.05$ 

**Table 6.** Species of *Fusarium* in kernels of winter wheat at Osiny in 2005–2010

				Proport	ion of ker	nels colo	nized [%]			
Taxon	0	I	С	M	mean	0	I	С	M	mean
			cv. Sukce	s				cv. Zyta	1	
			2005	5						
Fusarium avenaceum (Fr.) Sacc.	-	_	_	_	0.0	2.8	1.2	_	_	1.0
F. culmorum (W.G. Sm.) Sacc.	_	_	_	_	0.0	1.0	_	_	_	0.3
F. equiseti (Corda) Sacc.	-	_	_	_	0.0	_	_	1.0	_	0.3
F. oxysporum Schltdl.	_	_	_	_	0.0	_	_	1.2	_	0.3
F. poae (Peck) Wollenw.	2.2	1.0	6.5	4.0	3.4	2.0	7.8	4.0	7.5	5.3
F. sporotrichioides Sherb.	_	_	_	_	0.0	_	_	_	3.0	0.8
F. tricinctum (Corda) Sacc.	_	_	2.2	_	0.6	_	_	_	_	0.0
Fusarium spp.	2.2 bc	1.0 c	8.7a	4.0 b	4.0	5.8 b	9.0 ab	6.2 b	10.5 a	7.9
			2006	5						
F. avenaceum	1.2	_	_	_	0.3	_	_	_	_	0.0
F. langsethiae Torp & Nirenberg	_	-	_	_	0.0	_	1.5	-	_	0.4
F. роае	0.8	3.0	10.0	3.0	4.2	4.0	4.8	12.8	6.8	7.1
Fusarium spp.	2.0 b	3.0 b	10.0 a	3.0 b	4.5	4.0 b	6.3 b	12.8 a	6.8 b	7.5
			2007	7						
F. avenaceum	2.5	2.0	4.3	2.3	2.8	1.0	8.3	2.0	3.0	3.6
F. culmorum	-	1.5	2.0	_	0.9	_	1.0	3.3	_	1.1
F. graminearum	1.0	1.0	1.0	_	0.8	-	1.0	0.8	_	0.5
F. langsethiae	_	_	_	_	0.0	_	_	-	0.8	0.2
F. роае	3.8	12.5	6.0	18.2	10.1	14.0	10.0	18.0	17.5	14.9
F. sporotrichioides	_	-	0.8	0.8	0.4	1.5	_	4.5	_	1.5
F. tricinctum	2.0	7.8	4.8	4.0	4.7	4.3	7.0	4.0	8.5	6.0
Fusarium spp.	9.3 c	24.8 ab	18.8 b	25.3 a	19.6	20.8 b	27.2 ab	32.5 a	29.8 a	27.6
	cv. Lege	enda				cv. Ryw	ralka			
		- 10	2008							
F. avenaceum	_	1.0	1.8	_	0.7	1.5	1.5	1.0	0.2	1.1
F. culmorum	_	-	_	_	0.0	_	0.5	_	_	0.1
F. graminearum	-	1.0	-	_	0.3	_	-	-	_	0.0
F. poae	0.2	_	5.0	_	1.3	_	1.0	4.2	_	1.3
F. sporotrichioides	_	_	-	1.0	0.0	-	_	1.0	_	0.3
F. tricinctum	- 0.2	2.01	0.8	1.2	0.5	0.8	- 2.01	1.2	-	0.5
Fusarium spp.	0.2 c	2.0 b	7.6 a	1.2 bc	2.8	2.3 b	3.0 b	7.4 a	0.2 c	3.2
T grant general	1.0	4.0	2009		2.5	4.0	2.0	F 0	2.0	2 =
F. avenaceum F. culmorum	3.0	4.0	_	5.0 1.0	2.5 1.0	4.0	3.0	5.0	2.0	3.5 0.0
	2.0	2.0	2.0	1.0	1.0	2.0	_	5.0	1.0	2.0
F. graminearum F. poae		4.0	10.0	3.0	4.3	6.0	4.0	6.0	1.0	4.0
F. sporotrichioides	_	4.0	1.0	3.0	1.0	2.0	4.0	2.0	1.0	1.3
F. tricinctum	3.0	4.0	6.0	4.0	4.3	3.0	8.0	5.0	8.0	6.0
F. tricinctum Fusarium spp.	9.0 b	4.0 14.0 a	6.0 19.0 a	4.0 16.0 a	4.3 14.5	3.0 17.0 ab	8.0 15.0 b	5.0 23.0 a	8.0 12.0 b	16.8
Fusurium spp.	9.0 0	14.0 a	2010		14.3	17.0 ab	13.0 D	23.0 a	12.0 D	10.0
F. avenaceum	7.5	1.5	1.0	1.2	2.8	8.0	2.8		1.5	3.1
F. culmorum		1.5		1.2	0.0	2.5	4.0	_	1.5	0.6
F. graminearum	3.2	_	_	_	0.8	2.0	_	_	_	0.5
F. poae	1.5	1.2	4.2	5.0	3.0	2.8	_	2.0	2.2	1.8
F. tricinctum	2.0	1.0	4.2	1.0	1.0	2.0	_	2.0	Z.Z _	0.0
Fusarium spp.	2.0 14.2 a	3.7 c	5.2 bc	7.2 b	7.6	15.3 a	2.8 b	2.0 b	3.7 b	6.0
i nonituiti opp.	14.∠ a	J./ C	J.Z DC	7.4 D	7.0	10.0 a	۷.0 ک	Z.U D	5.7 0	0.0

O – organic, I – integrated, C – conventional, M – monoculture; a different letter in row indicates significant difference according to  $\chi^2$ -test at  $p \le 0.05$ 

2005 to 2010 (Table 5). When averaged over those 6 years, FDK ranged from 8.5% to 12.8%, and were generally the least in the organic system but with no significant differences between systems. In 2005 and 2008, a low amount of FDK (4.0-7.5% and 0.7-7.5%, respectively) was associated with a low FHB incidence. In 2005, the organic system had fewer FDK than other systems. In 2008, conventional cropping had higher FDK than other systems. In 2006, there was a moderate amount of the FDK (3.0-11.4%), despite the general absence of visible symptoms, and the FDK were significantly higher in the conventional than in other systems. In 2007 and 2009, the FDK were found to be higher than in other years, which was associated with moderate (2007) or low (2009) FHB incidence. In 2007, there were fewer FDK in the organic than in other systems. In 2009, there was more colonisation in the conventional than in other systems. In 2010, the FDK were exceptionally and significantly higher in the organic system than in other systems.

Fusarium poae was the Fusarium species most often isolated in the 2005–2010 period (Table 6). Fusarium poae was present every year, in each system, on each cultivar (with only a few exceptions). It colonised 1.3–14.9% of the kernels, on average, but locally up to 18.2% (i.e. cv. Sukces in 2007). Other Fusarium species occurred more rarely: F. avenaceum (Gibberella avenacea R.J. Cook) in 1.0–3.6% of the kernels, on average, F. culmorum in 0.1–1.1%, F. graminearum in 0.5–2.0%, F. sporotrichioides in 0.3–1.5%, and F. tricinctum (Gibberella tricincta El-Gholl, McRitchie, Schoult. & Ridings) in 0.5–6.0%. Fusarium equiseti (Gibberella intricans Wollenw.) and F. oxysporum occurred only in 2005, in the conventional system on cv. Zyta. Fusarium langsethiae occurred in 2006 and 2007, in 0.8 and 1.5% of kernels in the monoculture and

the integrated system, respectively. This was the first record of *F. langsethiae* in Poland (Łukanowski and Sadowski 2008; Łukanowski *et al.* 2008). Wheat kernels were also colonised by other fungi. The most common were *Alternaria alternata* (Fr.) Keissl. and *Epicoccum nigrum* Link.

Cultivation system affected the colonisation of kernels by individual Fusarium species in the 2005-2010 time period (Table 7). Kernel colonisation by F. poae was significantly different in each system. The most kernel colonisation by F. poae was in the conventional system and monoculture (355 and 269 kernels colonised, respectively, from 2400 evaluated), less in the integrated system (197 kernels), and the least in the organic system (149 kernels). Fusarium avenaceum occurred more often in the organic and integrated systems (118 and 101 kernels colonised) than in the conventional system and the monoculture (60 kernels). Significantly fewer kernels were colonised by F. tricinctum in the organic than in other systems. Fusarium culmorum and F. graminearum, the most important DON-producing FHB fungi worldwide, were relatively infrequent, but the former colonised more kernels in the organic system (26), and the latter infected more kernels in the organic and conventional systems (41 and 35) than in the other systems (statistically significant differences). Each of these fungi colonised the fewest kernels (4) in the monoculture. The cultivar did not affect the number of kernels colonised by most Fusarium species (Table 8). However, cv. Sukces was colonised significantly less than cv. Zyta by F. avenaceum, F. poae, and F. sporotrichioides.

There was a moderate positive correlation between FHB incidence and the proportion of FDK in each system in the 2005–2010 time period (r=0.616–0.790 at p ≤0.001) (Table 9).

Table 7. Species of Fusarium in kernels of winter wheat at different systems at Osiny in 2005–2010

System	Fusarium avenaceum	Fusarium culmorum	Fusarium equiseti	Fusarium graminearum	Fusarium langsethiae	Fusarium. oxysporum	Fusarium poae	Fusarium sporotrichioides	Fusarium tricinctum
			Number	of kernels c	olonized in 2	2,400 kernels	s sample		
Organic	118 a	26 a	0	41 a	0	0	149 d	14 b	60 b
Integrated	101 a	12 b	0	20 b	6	0	197 с	0 c	111 a
Conventional	60 b	21 ab	4	35 a	0	5	355 a	37 a	96 a
Monoculture	61 b	4 c	0	4 c	3	0	269 b	31 a	107 a

A different letter in a column indicates significant difference according to  $\chi^2$ -test at p  $\leq 0.05$ 

Table 8. Species of Fusarium in kernels of different cultivars of winter wheat at Osiny in 2005–2010

Cultivar	Fusarium avenaceum	Fusarium culmorum	Fusarium equiseti	Fusarium graminearum	Fusarium langsethiae	Fusarium oxysporum	Fusarium poae	Fusarium sporotrichioides	Fusarium tricinctum
			Number	of kernels c	olonized in 2	2,400 kernels	sample		
Legenda <sup>1</sup>	96 a	16 a	0	41 a	0	0	136 a	16 a	92 a
Rywalka <sup>1</sup>	122 a	12 a	0	40 a	0	0	113 a	24 a	104 a
Sukces <sup>2</sup>	49 b	14 a	0	12 a	0	0	284 b	6 b	83 a
Zyta <sup>2</sup>	73 a	21 a	4	7 a	9	5	437 a	36 a	95 a

<sup>&</sup>lt;sup>1</sup> in 2008–2010; <sup>2</sup> in 2005–2007; a different letter in a column indicates significant difference according to  $\chi^2$ -test at p  $\leq$  0.05

**Table 9.** Pearson's coefficients for Fusarium head blight (FHB) incidence and proportion of *Fusarium*-damaged kernels (FDK) in winter wheat at Osiny in 2005–2010

System	FHB	FDK	Linear regression coefficient <i>b</i>	Coefficient a	Correlation coefficient <i>r</i>	$\begin{array}{c} \text{Linear regression} \\ \text{equation } Y_p \end{array}$		
	Over all years, 2005–2010							
Organic	3.59	8.51	1.27	3.93	0.681	y = 1.27x + 3.93		
Integrated	4.46	9.32	1.18	4.06	0.732	y = 1.18x + 4.06		
Conventional	5.35	12.77	0.72	8.93	0.616	y = 0.72x + 8.93		
Monoculture	5.33	9.98	0.98	4.75	0.790	y = 0.98x + 4.75		
	Over all cropping systems							
2005	0.90	5.92	6.51	0.07	0.777	y = 6.51x + 0.07		
2006	0.00	5.99	_	_	_	_		
2007	15.5	23.56	0.7	12.68	0.629	y = 0.7x + 12.68		
2008	0.54	2.99	-3.0	4.6	-0.196	y = -3.0x + 4.6		
2009	3.1	15.62	-0.74	17.91	-0.288	y = -0.74x + 17.91		
2010	8.06	6.76	0.67	1.35	0.100	y = 0.67x + 1.35		
2005-2010	4.68	10.14	0.96	5.65	0.699	y = 0.96x + 5.65		

**Table 10.** Pearson's coefficients for the nitrogen content of extractable soil (NO<sub>3</sub> + NH<sub>4</sub>), phosphorus and potassium in the soil and Fusarium head blight (FHB) and *Fusarium*-damaged kernels (FDK) in winter wheat at Osiny in 2005–2010

Dependence	b	а	r	$F_{cal}/F_{tab}$	Correlation
Nitrogen – FHB	-0.07	13.7	-0.26	$F_{cal} > F_{tab}$	yes
Nitrogen – FDK	-0.10	18.0	-0.42	$F_{cal} > F_{tab}$	yes
Phosphorus – FHB	-0.57	15.1	-0.23	$F_{cal} < F_{tab}$	no
Phosphorus – FDK	0.31	6.3	0.15	$F_{cal} < F_{tab}$	no
Potassium – FHB	0.06	7.5	0.02	$F_{cal} < F_{tab}$	no
Potassium – FDK	0.32	6.7	0.13	$F_{cal} < F_{tab}$	no

 $F_{cal}$  – value calculated for ratio: explained variance/unexplained variance;

There was a moderate positive correlation between FHB incidence and FDK in two individual years; 2005 and 2007 (r = 0.777 and 0.629, respectively, at p  $\leq$  0.001).

An investigation was done on the relationship between the nitrogen content of extractable soil  $NO_3 + NH_{4'}$  phosphorus and potassium in the soil, and FHB and FDK. Correlation analysis showed only a small negative correlation between the nitrogen content of soil  $(NO_3 + NH_4)$  and FHB (coefficient r = -0.26) and between the nitrogen content and FDK (r = -0.42). No correlation was found between the content of phosphorus and potassium and FHB and FDK (Table 10).

## **Discussion**

Fusarium head blight is one of the major cereal diseases, being responsible for significant grain yield loss in wheat, barley, and oats. The disease is of additional concern because of mycotoxin production by the fungi involved, which poses a threat to the health of both human and animal consumers. The disease is highly linked to crop rotation, tillage, and weather conditions. Risk is considered to be particularly high in regions where: cereals form a large proportion of rotations, susceptible cultivars are grown, low-quality seeds are used, reduced or minimum tillage is applied (with debris left as a source of inoculum), and inappropriate sowing dates are chosen (Parry *et al.* 1995; McMullen *et al.* 1997; Dexter and Nowicki 2003; Champeil

et al. 2004; Klix 2008; Stein et al. 2009; Wegulo et al. 2011). Application of fungicides can reduce the damage level by 50–60% (under optimum conditions).

Manipulating the agricultural procedures can contribute greatly to reduced risk of disease, with less or no need for fungicides. More information is needed, however, on effects of the production system (including preceding and cover crops, tillage method, inorganic and organic fertilisation, and pesticide use) on disease.

Wheat grown after wheat in combination with minimal tillage, has been found to greatly increase the risk of FHB (Odorfer *et al.* 1994). In the present study, over the 9 year period, disease severity was low and mostly not significantly affected by a cultivation system. Fusarium head blight incidence, though, was often greater in the monoculture (wheat after wheat) than in other systems, and sometimes also in the integrated system, although minimum tillage was not applied.

It may seem that wheat grown in organic systems, without the protection of chemical fungicides, would be more susceptible to infection and colonisation by *Fusarium*. Yet, the average FHB incidence was not greater in the organic system, except in 2009 when disease was slight (statistically higher). This may result partly from environmental conditions created by the smaller number of plants growing in the organic system (100–150 heads · m<sup>-2</sup>), despite the similar amounts of seed sown (Kuś *et al.* 2010). The lower density of plants helps to create a specific

 $F_{tab}$  – critical value for significance level 0.05



microclimate; with lower humidity, higher temperatures, and more exposure to sun and wind. Such a microclimate increases the development of kernels, helps the heads to dry and decreases the time of their exposure to *Fusarium* infection. In different cultivation systems, similar FHB incidence has been reported by Champeil *et al.* (2004), Sadowski *et al.* (2008) and Lenc *et al.* (2011). Seasonal differences in relative amounts of FHB in different systems, especially the occasionally higher levels in the organic and integrated systems, may have resulted from the unrecognised effects of the treatments. This includes such effects as the residues of different cover crops (see below).

Correlations have usually been found between FHB incidence, proportion of FDK and DON content. Yet, in some wheat cultivars no correlation or negative correlations have been reported (Mesterházy et al. 1999, 2003; Bai et al. 2001; Bai and Skaner 2004; Koch et al. 2006; Brennan et al. 2007; Yuen and Shoneweiss 2007; Lehoczki-Krsjak et al. 2010; Wegulo et al. 2011). The grain from Osiny was colonised by mycotoxin-producing Fusarium spp., the most frequent of which, F. poae, is known to produce a wide range of toxins (e.g. Steinglein 2009). The grain was not assessed for DON or other mycotoxins, but FHB incidence was moderately correlated with FDK and the extent of disease incidence. Kernel colonisation by Fusarium spp. suggests that mycotoxins may have been present. The cropping system did not usually significantly affect FDK, but FDK tended to be least in the organic system. This suggests that wheat grain from the organic system may have contained less mycotoxin than grain from other systems. This is in agreement with Birzele et al. (2002), Champeil et al. (2004), Schneweis et al. (2005), and Bernhoft et al. (2010). Nevertheless, Edwards (2006) found no significant differences in DON content between wheat grain grown organically and conventionally.

In many areas, fungicides are rarely used for FHB control because of the fungicides' high cost, variable efficacy, and also because of the erratic nature of FHB epidemics. Standard fungicide treatment applied at the first stages of stem elongation (GS 31-33) and inflorescence emergence (GS 51-55), usually provides more successful protection against non-Fusarium species than Fusarium species. Better protection can be provided with fungicide treatment applied at flowering (GS 60-70), shortly before, during or after infection (Sirranidou and Buchenauer 2000; Lenc et al. 2009; Sadowski et al. 2009, 2011). Mielke et al. (2000) reported a 67% effectiveness of fungicides in FHB control when spraying is done 3 days before infection. Only a 35% effectiveness was found when spraying as done 3 days after infection. In the present study, fungicides were usually applied twice (integrated system) or three times (conventional system and monoculture), starting at the end of April, through May to June (at booting, inflorescence emergence, and the flowering stages, GS 40-60). Two fungicide formulations containing tebuconazole, known for their effectiveness in the control of Fusarium diseases, were used. Tebuconazole, when applied at flowering of wheat, can reduce FHB incidence by about 60-65%, increase grain yield, and reduce the mycotoxin content (Birzele et al. 2002; Gromadzka et al. 2012). In the present study, however, fungicide treatment did not appear to contribute to significant FHB control when compared with the non-treated organically grown crop. If fungicides were effective, then they were generally less effective than the alternative procedures applied in the organic system. Lower FHB severity could affect a smaller density of shoots on the field, arising for example, from a weaker tillering of plants, which usually occurs in the organic cultivation. Shoot density affects the field moisture.

Fusarium head blight also occurs on other small grain crops and corn. If used in rotation these plants provide a continuous availability of hosts and increase the potential for carry-over of the pathogen into subsequent seasons. Rotation must provide time for the residues to break down and the pathogen population to decline (Parry et al. 1995; Klix et al. 2008). Fusarium head blight incidence is usually less in wheat grown after legumes. Therefore, the Osiny experiment included legumes commonly grown in central Europe (clover, faba bean, lupin, and common vetch) as preceding crops or cover crops in the organic and integrated systems. The wide selection of legumes and their extended application (for 2 years as preceding crops) may have contributed to a lesser FHB incidence and severity, occurring in the organic system. Oilseed rape was the only preceding crop used in the conventional system but was rarely associated with the highest rate of FHB incidence. Earlier reports of oilseed rape use describe contradictory effects. Fernandez et al. (2005) reported higher FHB incidence, while Sadowski et al. (2011) reported a similar FHB incidence and kernel colonisation in wheat following oilseed crops, when compared with wheat following wheat.

Rotation alone is not sufficient to prevent disease. Generally, tillage to incorporate crop residues into the soil (i.e. ploughing) can significantly reduce the risk, even when cereal was the previous crop, by burying the inoculum source (Parry et al. 1995; Klix et al. 2008). Buried wheat residues decompose faster than those on the soil surface. After 2 years, only 2% of buried residues remain, in contrast to 25% of the residues on the soil surface. In the present study, the more intensive tillage in the integrated system, compared with that in the conventional system, did not decrease the FHB incidence and sometimes tended to increase it. It is possible that a second ploughing in the integrated system had the effect of returning buried crop residue to the surface. Such an effect was not apparent in the similarly treated organic system, which was apparently less conducive to FHB for other reasons.

Growing cereal cultivars with reduced susceptibility to FHB is the most promising strategy for disease control because of its potential and low costs. The present results show significant differences within pairs of wheat cultivars on FHB incidence in two years (2002, 2004) out of three (2002, 2004, 2007) in which FHB incidence reached > 10%. Fusarium head blight incidence was greater on cv. Juma than Elena in 2002, and on cv. Zyta than Sukces in 2004. These differences were not maintained for all the years of the study, but were partly supported by *Fusarium* colonisation results (from 2005–2010), which showed that cv. Sukces was often significantly less colonised than cv. Zyta by the most common *Fusarium* spp. Other reports have shown



the significant effects of wheat cultivar on the colonisation of kernels by *Fusarium* spp. (Klix *et al.* 2008; Lenc *et al.* 2009; Lenc 2011). Despite considerable research effort, since 1990 no breeding lines offering complete resistance to FHB have been identified (Kollers *et al.* 2013). The cultivars tested here, due to their relatively high infection do not seem to be good candidates for resistance breeding.

Weather (especially rainfall before and after flowering) was the main determinant of FHB incidence at Osiny. Fusarium head blight incidence was low in 2005, 2006, and 2008 when rainfall in June was least, and higher when rainfall was high and more prolonged. Higher humidity favours the growth of FHB fungi and spread of disease while lower humidity helps keep plants dryer and prevents infection and colonisation. Others have also reported the dependence of FHB incidence on the weather (Parry *et al.* 1995; Jennings and Turner 1996; Kiecana *et al.* 1997; De Wolf *et al.* 2003; Lemmens *et al.* 2004; Xu *et al.* 2008; Cowger *et al.* 2009).

Diversity of Fusarium species in colonised wheat kernels is mostly the result of region and climate. There was generally a low FHB incidence at Osiny in 2005-2010, when kernels were colonised predominantly by F. poae. This species, together with F. avenaceum, F. culmorum, 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, the USA, and Canada (Wilcoxon et al. 1988; Gale 2003; Gale et al. 2007; Xu and Nicholson 2009; Alvarez et al. 2010). In years with the most FHB, F. poae was accompanied by F. avenaceum and F. tricinctum. They were usually equally distributed among cultivars. Only F. langsethiae, F. oxysporum, and F. tricinctum occurred just on the Zyta cultivar. The marginal presence of F. culmorum and F. graminearum, moderate presence of F. avenaceum, and absence of M. nivale was unexpected. The last three were dominant species in Flanders, in the temperate zone, which has a similar climate to that of Poland (Audenaert et al. 2009). Local dominance of *F. poae* in the FHB populations and the lack of a wheat cultivar effect on local FHB population diversity was also recorded in Flanders (Audenaert et al. 2009). The composition of the FHB populations could, however, have been influenced by weather conditions. Rintelen (1995) observed that if the weather does not favour the development of strongly toxigenic F. culmorum and F. graminearum at the flowering stage of wheat, it may favour the later development of *F. poae* and *F. tricinctum*.

## Conclusion

Fusarium head blight, with *F. poae* as the dominant pathogen, was most frequent where rainfall was high and prolonged during the flowering and ripening of wheat. Fusarium head blight tended to appear the least in the organic system and was the most in monoculture, and sometimes, the integrated system. The differences may be due to the crop sequences, amounts of inversion tillage, and preceding or cover crops.

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