Contamination of winter wheat with *Fusarium* mycotoxin depending on environment conditions and cultivar

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Abstract. The objectives was to investigate relationship between weather conditions, cultivar and deoxynivalenol (DON) concentration in winter wheat. Concentrations of DON were analyzed in 10 winter wheat cultivars: Batuta, Ostroga, Markiza, Nateja, Muszelka, Tonacja, Izyda, Satyna, Fregata, Kohelia. The winter wheat cultivars were naturally contaminated and collected from different locations of Poland, i.e. Srem Wojtostwo (φ = 52o05′, λ = 17o02), Radostowo (φ = 53o59′, λ = 18o45), Czeslawice (φ = 51o19′, λ = 22o16′). Grain samples were analyzed for contamination with deoxynivalenol (DON) by the enzyme-linked immunosorbent analysis (ELISA) method. The cultivar and weather conditions, as well as field locations strongly modified the concentration of deoxynivalenol in grains. The samples of Tonacja, Ostroga and Muszelka cultivars showed higher contamination with DON, but the lowest Nateja, Kohelia, Batuta cv. The precipitation in May, June and July strongly affected seed contamination. More rain contributed to increased wheat contamination with DON.

Keywords: winter wheat, cultivar, mycotoxin, deoxynivalenol, weather conditions, morphological features.

1 Introduction

The occurrence of mycotoxins in various crops is a food safety issue of a great concern worldwide. Mycotoxins that are frequently found in cereals are secondary metabolites produced mainly by *Fusarium* ear blight pathogens, which are common in the temperate climatic zone of Europe, America, and Asia (Creppy et al. 2002). None of the geographical regions is free from the occurrence of mycotoxins. Contamination with these compounds affects 25 percent of the world crops annually. Economic costs incurred by crop producers due to mycotoxin contamination of the grains of cereals, oilseeds, and feed are estimated at nearly billion dollars a year. The particularly dangerous and commonly occurring is deoxynivalenol (DON) mainly produced by *Fusarium culmorum* and *Fusarium graminearum* (Bryden, 2012, Creppy et al. 2002, Hajšlová et al. 2007, Marin et al. 2013).

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The occurrence of Fusarium mycotoxins, mainly DON, in cereal grains as well as in various cereal based foodstuffs has been reported in several studies. The report (Schothorst and Egmond 2004) provides an overview of Fusarium toxin levels in crops harvested in 12 European countries (Austria, Belgium, Denmark, Finland, France, Germany, Italy, the Netherlands, Norway, Portugal, Sweden, the United Kingdom). DON was found in many samples. Similarly, the survey presented by Joint FAO/WHO Expert Committee on Food Additives (JECFA 2001) showed that DON was the most abundant mycotoxin in cereals, regardless of the country they were harvested in. In southwest Germany, the presence of DON in wheat flour was detected in 98% samples. DON content was in the range from 15 to 965 μg kg^{-1} in white flour, and from 15 to 1379 μg kg^{-1} in wholegrain flour (Schollenberger et al. 2002). Also in Poland, the cyclical contamination of cereal grains with DON had been observed. (Cegielska-Radziejewska et al. 2009). The incidence of mycotoxins can vary from year to year depending on many factors such as weather conditions and/or agricultural practices (Obst et al., 2000). Edwards (2004), Czaban et al. (2015), Mikos and Podolska (2013) reported that besides weather conditions, a cultivar had an important role in mycotoxin contamination of cereal kernels. The objective of this research was to investigate relationship between weather, environment conditions, cultivar and DON concentration in winter wheat.

2 Materials and methods

2.1 Field experiments

The experiment was carried out in the years of 2009-2011 in 3 Experimental Stations located in different regions of Poland: Srem Wojtostwo (φ = 52o05´, λ = 17o02´) , Radostowo (φ = 53o59´, λ = 18o45´), Czeslawice (φ = 51o19´, λ = 22o16´). The experiment involved 10 winter wheat cultivars: Batuta, Ostroga, Markiza, Nateja, Muszelka, Tonacja, Izyda, Satyna, Fregata, Kohelia. The cultivars differed from each other in terms of heading, wax coating on head, plant height, stem pith, density of spike, length of spike, arista, length of arista, width of lemma, shape of lemma, length of lemma. The crop managements (sowing term, sowing, density, fertilization, crop protection were on the same level in each experimental stations). The winter wheat was sown on very good (Radostowo) and good wheat soil complexes (Srem Wojtostwo, Czeslawice). The winter wheat was harvested in full maturity phase. In each experimental stations, during vegetation period of winter wheat, weather conditions (temperature, precipitation) were monitored. Grain samples were analyzed for DON contamination with toxins. Preparation of the grain samples and ELISA test procedure for determination of contents of deoxynivalenol (DON) in the grain were carried out according to Cegielska-Radziejewska et al. (2009), Mikos-Szymanska and Podolska (2013), Czaban et al. (2015).

Analysis of mycotoxin contents: a quantitative analysis of DON was carried out using enzyme-linked immunosorbent analysis (ELISA) commercial kit (Neogen Corporation, Food Safety Diagnostics: Veratox® for DON 5/5 – 8331NE). The

method is based on the antibody-antigen interaction and has been approved by the AOAC Research Institute (Certificate No. 950702).

2.2 Statistical analysis:

Statistica 6.0 software (StatSoft Inc.) was used for statistical analysis. The compatibility of variables with normal distribution was tested using the Shapiro-Wilk test. Verification of the hypothesis was performed using nonparametric (Kruskal-Wallis test). Assumed significance level = 0.05.

3 Results

The applied treatments (year, location and cultivar) modified the winter wheat mycotoxins contamination by DON.

Concentration of mycotoxin in winter wheat seeds significantly depended on the vegetation season (years). In all years, the investigated mycotoxins were detected, but at different levels. The highest DON concentration was observed in 2011 year, lower in 2010, but the lowest in 2009 (Table 1). The concentration of DON in winter wheat seeds from Srem Wojtostwo was 10,0 μg kg^{-1}, from Radostowo 146,7 μg kg^{-1}, while from Czeslawice 273,3 μg kg^{-1} (Table 1).

In the present research, a high correlation between concentration of mycotoxin and precipitation has been found (Table 2). The occurrence of DON was similarly affected by rainfall in May (r = 0.89819), June (r = 0.80220), July (r = 0.84016). The number of DON was not significantly correlated with temperature in May, June and July (Table. 2).

Table 1. DON contamination (μg kg^{-1}) of winter wheat seeds depending on the years and localization

Year	2009	2010	2011	Śrem Wojtostwo	Radostowo	Czesławice
DON	86, 7	106,33	256, 7	10,0	146,7	273,3

Table 2. Correlation coefficient and simple regression between DON contamination of winter wheat seeds and precipitation and temperature (data from 2009-2011).

Month	Precipitation		Temperature	
	Correlation	Correlation coefficient	Correlation	Correlation coefficient
May	=-0,3255+0,01379x	r= **0,89919**	= 1,5746 - 0,0962x	r= -0,3926
June	=-0,3139+0,01045x	r= **0,80220**	= 0,66670 - ,0216x	r= -0,0946
July	=-0,2721+0,00396x	r= **0,84016**	= 0,42167 – 0,080x	r= -0,0176

bold-significant correlation

The contamination of winter wheat cultivars in 2011 was shown in Figure 1. There were significant differences cultivars in terms of DON contamination in 2011. The concentration of DON ranged from 200 μg kg^{-1} (Nateja cv.) to 1400 μg kg^{-1} (Tonacja cv.). Significantly the lowest concentration of DON was recorded for the seeds Nateja cv. compared with Tonacja, Muszelka and Fregata. The concentration of DON was significantly lower in the seeds of Nateja, Satyna and Batuta.

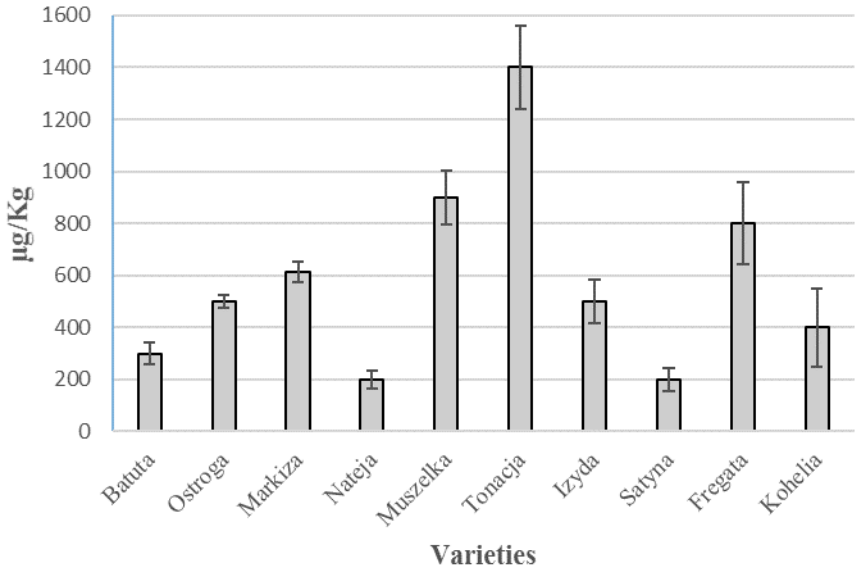

Figure 1. Concentration of DON in winter wheat cultivars (2011 year).

In order to find correlation between morphological characteristics of winter wheat cultivars and mycotoxin contamination, eleven features were taken into consideration There was a significant negative correlation between plant height and stem pith. Shorter cultivars with thin stem pith were the most contaminated. (Table 3).

Table 3. Correlation coefficient between morphological characteristics of winter wheat cultivars and mycotoxin contamination

1	2	3	4	5	6	7	8	9	10	11
-0.04	0.39	**-0.71**	**-0.58**	0.12	0.48	0.18	0.17	0.47	0.48	0.24

bold- significant correlation

(1-term of heading, 2-wax coating on head, 3-plant high, 4-stem pith, 5-dense of spike, 6-length of spike, 7-arista, 8-length of arista, 9-with of lemma, 10-shape of lemma, 11-length of lemma).

4 Discussion and Conclusions

Research conducted in numerous countries has shown that grain contamination by Fusarium and mycotoxin level were affected mainly by weather conditions (Bryla et al. 2016). Moderate temperatures (15-30°C) combined with prolonged periods of high humidity during the blossoming and/or earing phases promote the accumulation of DON and are the best indicators of fusariosis (Hooker et al..2002, Cowger et al. 2009.). Bernhoft et al. (2012) reported that low temperature before harvest increased DON concentration in wheat grain. Favourable conditions – high precipitations and relatively high temperatures during the period when the wheat is most susceptible to infection (i.e. from flowering to the soft dough stage of kernel development) (Edwards 2004, Obst et al. 2000 Hajslova et al., 2007) were obviously factors responsible for the high mycotoxin levels. It has been confirmed in the present study. We found a high correlation between concentration DON and precipitation in May, June and July. More rain contributed to increased wheat contamination with DON.

We also found that besides weather conditions, cultivars play important role in DON concentration in winter wheat. The significantly lowest concentration of DON was noted in the seeds of Nateja, Kohelia, Batuta cv. compared with Tonacja, Ostroga and Muszelka. Similar results were obtained by Bryla et al. (2016). Among 10 cultivars, Fidelius cv. was the least contaminated with DON, while Bamberka Forkida and Kampana, the most (Bryla et al. 2016). Bai et al. (2001) who conducted research in artificial inoculation of winter wheat by *F. graminearum*, proved significant differences in DON level among cultivars. Out of 116 varieties, they selected 16 with a small amount of DON. They found that these varieties may be useful as sources for breeding wheat cultivars with lower DON level. They also found that cultivars with scabbed spikelets and scabbed kernels had less level in DON. In our study we found that shorter winter wheat cultivars and cultivars with thin stem pith were the most contaminated with DON.

Our studies have confirmed a significant impact of weather on the amount of DON in wheat kernels, and indicated that, under conditions conducive to the formation of toxins, there are varieties that accumulate small amounts of mycotoxins. Such varieties should be recommended for cultivation as safe for health reasons.

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