EFFECT OF DIFFERENT INTENSITIES OF *FUSARIUM* INFESTATION ON GRAIN YIELD, DEOXYNIVALENOL CONTENT AND BAKING QUALITY OF WINTER WHEAT

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ABSTRACT

The effects of different infection grades of *Fusarium* spp. on grain yield, deoxynivalenol (DON) content and baking quality of winter wheat were evaluated in 3 wheat cultivars (Akteur, Eurofit and Meritto) during the 3 years field trials (2008 - 2010), where different ways of artificial inoculation and protective fungicidal treatment (8 variants) were used to obtain a scale of wheat samples with different levels of *Fusarium* spp. infection (determined using the PCR assay).

Inoculated variants with the highest *Fusarisum* infection grade also reached the highest DON content (determined using the method based on UPLC-TOF MS system) and exceeded considerably the limit of DON content for the food grain (1250 µg.kg⁻¹). Significant differences in the grain yield were estimated between the inoculated variants and the check variant. Effect of fungicidal treatment against *Fusarium* spp. was positive and led to decrease of DON content and increase of the grain yield.

Although the crude protein content and wet gluten content were not significantly affected by the *Fusarium* spp. infection, ZS (Zeleny sedimentation) and FN (falling number) showed distinctively decreased values in the inoculated variants. Increasing intensity of *Fusarium* spp. infection evidently worsened the rheological quality, evaluated using the Mixolab system and a negative effect on protein and mainly on the starch part of the grain was obvious from Mixolab characteristics. High correlations were found between Mixolab characteristics and standard technological quality parameters, as well as between Mixolab characteristics and the main baking criterion – loaf volume of bread. Protective fungicidal treatment had a positive effect and led to improved values of most evaluated baking quality parameters.

Key words: wheat, Fusarium head blight, deoxynivalenol, grain yield, baking quality, Mixolab.

INTRODUCTION

P athogen fungi *Fusarium* spp. are the cause of *Fusarium* head blight (FHB), a serious threat to the worldwide grain industry in favorable conditions for development of epidemics. The *Fusarium* spores are disseminated by wind under favourable weather conditions to wheat spikes at flowering time (Mauler-Machnik and Suty, 1997).

FHB induces, at first, a premature fading of individual spikelets and then, subsequently, a fading of the whole ears. This can result in serious yield losses and a degradation of wheat quality (Eggert et al., 2010). Boyacioglu and Hettiarachchy (1995) characterized *F. graminearum* as an aggresive invader destroying starch granules, storage proteins and cell walls. According to Dexter et al. (1996) *F. graminearum* infection can cause significant changes in carbohydrate, lipid and protein composition. Additional changes also seriously affected the quality of storage proteins and dough properties in Canadian red hard spring wheat.

Furthermore, an outbreak of FHB is often accompanied by mycotoxin contamination, such as the very stable trichothecene deoxynivalenol, which is not degradated during storage, milling, processing or cooking of food, nor during treatment at high temperatures (Wang et al., 2005). DON is considered as a contamination marker subjected to the European Commission Regulation (EC 2006). DON maximum level was set at 1250 μ g.kg⁻¹ for unprocessed cereal grain and 750 μ g.kg⁻¹ for cereal flours. There are contradictory reports in the literature concerning the close correlations between FHB and DON content, but it is accepted that overall, accumulation of DON in kernel also would require successful infection and colonization stages of host (Smith et al., 2004).

The increased intensity of FHB over the last years has been attributed to the widespread adoption of minimum tillage and unsuitable crop rotation with many host crops. Tillage and crop rotation with a broad scale of crops are management strategies often recommended reduce residue-borne to inoculum density and pathogen survival. suggest Many researches that high concentration of fungus spores can be found under conditions with high inoculum density (Váňová et al., 2008). Agricultural measures like balanced crop rotation of cereals and noncereal crops, avoidance of maize as a preceding crop and soil tillage may contribute to the reduction in the risk of FHB disease and toxin contamination (Matthies and Buchenauer, 2000). Besides these measures and choosing of the more resistant cultivars, a direct measure, consisting in chemical control may contribute to the reduction in the risk of FHB disease and toxin contamination. More effective fungicides against FHB are always developed (Mesterházy et al., 2003).

Despite the fact, that some authors mentioned negative effects of FHB on breadmaking quality of wheat and the reduction of loaf bread volumes (Nightingale et al., 1999; Gärtner et al., 2008), there are also some contradictory studies, where a Fusarium infection strong did not significantly influence the breadmaking properties (Antes et al., 2001; Prange et al., 2005).

The objective of this study was to determine the effect of different infection grades of *Fusarium* spp. on grain yield, DON content and breadmaking quality of winter

wheat and to detect the changes in the breadmaking quality of winter wheat due to different levels of *Fusarium* spp. infection, using the new rheological system Mixolab.

MATERIAL AND METHODS

Plant material

A set of winter wheat samples from the exact field plot trial, conducted in the years 2008, 2009 and 2010 on the experimental station of the Department of Crop Production of the Czech University of Life Sciences Prague in Uhříněves (295 m above sea level, average annual temperature 8.4°C, average sum of precipitation 575 mm) was used for evaluation of the effect of different FHB infestation intensities on grain vield. deoxynivalenol content in grain and baking quality parameters. Field plot trials were carried out by the method of randomized blocks in three replications; average size of experimental plot was 12 m². The set of samples included 3 winter wheat cultivars (Akteur – quality group E; Eurofit – quality group A; Meritto – quality group B) and 8 field plot treatments (Table 1).

Artificial inoculation of wheat and evaluation of FHB infestation

Different ways of artificial inoculation of wheat and simultaneously several ways of fungicide treatment were used in the trials, with the aim to obtain a scale of wheat samples with different levels of *Fusarium* spp. infection (Table 1).

The isolates of F. culmorum and F. graminearum used for the artificial obtained inoculation were from the mycological collection of the Crop Research Institute in Prague and cultivated on sterile wheat grains. Wheat grains with the cultures of F. culmorum and F. graminearum were put into a vessel with water and shaken for 15 min in a laboratory shaker to release the spores into water. The obtained suspension was filtered through the gauze. Then artificial inoculation was made with the suspension of F. culmorum and F. graminearum spores in the ratio of 1:1, 10^7 spores.ml⁻¹ (Bürker chamber was used for the verification of

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inoculums density), 2 1 of suspension per experimental plot (12 m^2) . The suspension was dosed according to the list of variants with a hand sprayer at the beginning and at the end of the wheat flowering. The fungicide Prosaro (effective substances prothioconazole + tebuconazole) was applied two days before the inoculation.

Variant signif.	Treatment									
Ι	Check variant without any treatment									
II	Protective fungicidal treatment									
Ш	Artificial inoculation at the beginning of wheat									
111	flowering									
IV	Artificial inoculation at the beginning of wheat									
1 V	flowering + fungicide									
V	Artificial inoculation at the end of wheat									
v	flowering									
VI	Artificial inoculation at the end of wheat									
V I	flowering + fungicide									
VII	Artificial inoculation at the beginning and at									
VII	the end of flowering									
VIII	Artificial inoculation at the beginning and at									
v III	the end of flowering + fungicide									

PCR assay was used for *Fusarium* spp. detection. Total DNA was extracted from the grain samples using the DNeasy Plant Mini Kit (Qiagen, Hilde, Germany). The sequences of the primers, PCR reaction mixture and the conditions used for detecting of *F. culmorum* and *F. graminearum* in this study were identical to those described by Schilling et al. (1996).

DON determination

Analytical method based on UPLC-TOF MS system was used for DON analyses. Analytical standards of DON were obtained from Biopure (Austria). All of wheat samples were milled and homogenized prior to extraction procedure. Representative sample (12.5 g) was extracted with 50 ml of mixture of acetonitrile: water (84:16, v/v) for 60 min. Aliquot of crude extract (4 ml) was evaporated to dryness and the residue was then dissolved in 1 ml of methanol: water (1:1,v/v) mixture. Separation and quantification of analytes were carried out by

UPLC-TOF MS system consisting of Ultra-Performance Liquid Chromatography (Acquity, Waters, USA) coupled to a time-offlight mass spectrometer (LCT Premier, Waters, USA). For separation of analytes the Acquity UPLC HSS T3 column (100 x 2.1 mm I.D., 1.7 µm particle size, Waters, USA) was used. The fast linear gradient program for separation of target compounds was used. The orthogonal time-of-flight mass spectrometer operated in negative electrospray was ionization (ESI) mode. Capillary voltage was established on 3500 V, cone voltage 40 V, source temperature 120°C, desolvation temperature 350°C. Nitrogen was used as a desolvation as well as cone gas. Target analytes were identified according to their retention times and accurate masses. Software MassLynx 4.1 with application on QuanLynx manager was used for data acquisition and processing.

Standard technological quality parameters

Within the frame of the baking quality, crude protein content (CP) according to the Kjeldahl method (EN ISO 20483; ICC-Standard No. 105/2) and wet gluten content (WG) in grain dry matter using the apparatus Glutomatic Perten (ISO 5531), falling number (FN) – ISO 3093, Zeleny sedimentation index (ZS) – ISO 5529, ash content (AC) – ISO 2171 and volume weigh (VW) – ISO 7971-2 were determined.

Mixolab determination

The wheat grain samples were milled on a laboratory Bühler mill automat MLU 202. All milling fractions (3 fractions of break flour and 3 fractions of reduction flour) were collected (total flour yield 65% approximately) and used for the evaluation of the rheological properties and for the baking trial.

Rheological characteristics were determined using the apparatus Mixolab (Chopin, Tripette and Renaud, Paris, France) according to the Mixolab protocol Chopin + for the white flour (Mixolab appl., Handbook, 2008). Mixolab records in real time the torque (in nm) produced by the dough between two blades. A typical Mixolab curve is separated into five stages, characterised by the five points (C1-C5) and other parameters resulting from the differences between the individual points (Figure 1). Characteristics evaluated from the measured Mixolab curve are:

C1 - used to determine water absorption;

C2 - represents the weakening of the protein based on the mechanical work and the increasing temperature;

C3 - represents the rate of starch gelatinisation;

C4 - represents the stability of the hotformed gel;

C5 - represents starch retrogradation during the cooling period;

C1 - C2 - indicates the protein network strenght under increasing heating;

C3 - C4 - shows diastatic activity and relates with falling number;

C5 - C4 - correlates with the anti-staling effects, represents the shelf life of the end products.

Dough stability indicates the stability of the dough before weakening.

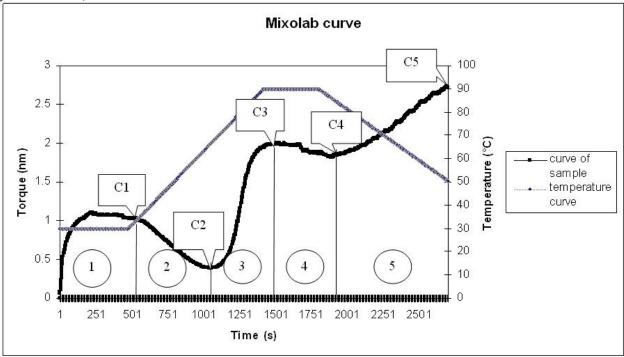


Figure 1. Standard Mixolab curve

The first two stages of the curve correspond to the rheological characteristics of stability, elasticity and water absorption, related to proteins. The consistency of dough is an indication of protein weakening. The faster and greater is the decrease of the dough consistency, the lower is the protein quality. The other stages relate mainly to starch and amylolytic quality. The increase in the consistency curve is mainly due to starch gelatinisation. At the fourth stage, consistency decreases as a result of fading amylolytic activity and bursting of starch granules. At the fifth stage, there is a decrease in the temperature and an increase in consistency as a result of the gel formation. This stage depends mainly on the level of amylose present, which starts the process of starch retrogradation (Rosell et al., 2007).

Baking trial

Bread was made according to the internal laboratory protocol from flour (300 g), yeast (12 g), fat (3 g), sugar (4.5 g) and salt (5.1 g). The dough was kneaded using a farinograph with the water addition according to the determined retention capacity of the flour. The dough heaved in thermostat for 45 min in 30° C and then it was divided to four ballshaped parts and left in a thermostat for 50 min. Afterwards the pieces were put in the oven at 240°C for 14 min. After cooling (90 min), baking characteristics were measured, i.e. the height (H) and diameter (D) of breads, and the ratios H/D were calculated. The loaf volumes (LV) were determined by rapeseed displacement.

Statistical analysis

The results were statistically evaluated by the analysis of variance (ANOVA) with subsequent Tukey HSD test. Their relations were assessed by the correlation analysis with the statistical significance expression on the level $\alpha = 0.05$; 0.01. The calculation was done using the software STATISTICA 8.0 CZ (StatSoft).

RESULTS AND DISCUSSION

Intensity of *Fusarium* spp. infestation, DON content and grain yield

According to the expectation, the highest intensity of Fusarium spp. infection was determined in variant with artificial inoculation at beginning of flowering (III) and in variant with artificial inoculation at beginning and at the end of flowering (VII) (Table 2).

	Cultivar	2008		2	2009	2010		
Variant		<i>F</i> .	<i>F</i> .	<i>F</i> .	<i>F</i> .	<i>F</i> .	<i>F</i> .	
		culmorum	graminearum	culmorum	graminearum	culmorum	graminearum	
	Meritto	++	+	+	+	-	-	
Ι	Akteur	++	-	+	-	-	-	
	Eurofit	++	-	-	+	-	-	
	Meritto	+	-	+	-	-	-	
II	Akteur	+	-	+	-	-	-	
	Eurofit	-	-	+	-	-	-	
	Meritto	+++	+	++	+	+++	-	
III	Akteur	+++	+	++	+	++	++	
	Eurofit	++	+	++	+	++	-	
	Meritto	++	-	+	-	+	+	
IV	Akteur	++	-	+	-	+	+	
	Eurofit	++	-	+	-	++	+	
	Meritto	+	-	+	-	++	-	
V	Akteur	+++	+	++	-	++	++	
	Eurofit	+	-	+	-	+	+	
	Meritto	++	-	+	-	++	-	
VI	Akteur	++	+	+	-	++	-	
	Eurofit	++	-	-	-	++	-	
	Meritto	+++	+	++	+	+++	+	
VII	Akteur	+++	+	++	+	++	+	
Ē	Eurofit	++	-	++	-	++	+	
	Meritto	++	+	+	-	++	-	
VIII	Akteur	++	-	+	-	++	+	
	Eurofit	++	+	+	-	++	-	

Table 2. Fusarium infection grade of the wheat samples (PCR assay)

+ sample positive (low infection grade); ++ sample positive (medium infection grade);

+++ sample positive (high infection grade); - sample negative.

Although suspension of *F. culmorum* and *F. graminearum* spores in the rate 1:1 was used for artificial inoculation, *F. culmorum* predominated markedly on the resulting grain infestation. So, results of the grain infestation did not reflect percentage composition of the used inoculator. These results are in accordance with findings of Prange et al.

(2005), who used for artificial inoculation of wheat a "cocktail" compounded from four species of *Fusarium* and determined considerable differences in resultant representation of these species on the inoculated wheat. It is also evident from our results that *F. culmorum* predominated in variants without artificial inoculation, as well.

According to the current knowledge. F. graminearum is prevalent in warmer regions, while F. culmorum predominates in the cooler climates of north-western Europe (Parry et al., 1995). According to our other findings, protective fungicidal treatment by Prosaro led in most cases to reduction of Fusarium spp. infection grade (Table 2). The differences are visible between the individual years too. In 2010 and especially 2008, the infection grade was evidently higher than in 2009. It was most likely due to the climatic conditions during the wheat flowering, which were favourable for a fast development of Fusarium spp. in these years. It is evident from our results (Table 3), that the DON content in the wheat grain samples varied

widely. in dependence on experimental Inoculated variant. variants (especially variants III and VII) with the highest Fusarium infection grade, reached also the highest DON content and exceeded considerably limit of DON content for the µg.kg⁻¹). food grain (1250 Fungicidal treatment by Prosaro proved a positive effect and markedly decreased DON content in the wheat grain, as seen from the differences between the variants III and IV and between the variants VII and VIII (Table 3). Řehořová et al. (2008) reached similar results – according to their findings the effect of fungicidal treatment against Fusarium spp. was very positive and led to decreasing of DON content in grain by more than about 50%.

Table 3. Average values of DON content in grain, grain yield, TKW and VW for individual variants, years and wheat cultivars

Sussification	DON	Grain yield	TKW	VW
Specification	$(\mu g.kg^{-1})$	$(t.ha^{-1})$	(g)	$(kg.hl^{-1})$
Ι	108.6ab	7.62ad	44.88ac	75.67a
II	27.1a	7.96a	45.71a	77.60a
III	3135.3e	6.12b	39.91bd	69.18b
IV	1496.8c	6.96c	42.51bc	71.01b
V	1814.0d	6.85c	41.97bd	69.88b
VI	338.3b	7.45de	43.66ac	72.58b
VII	3886.4f	5.95b	38.87d	65.62c
VIII	1680.8cd	7.04ce	43.26ac	70.23b
2008	2317.4c	7.02a	40.49b	68.01c
2009	639.2a	7.69b	44.04a	74.73b
2010	1529.8b	6.25c	43.25a	71.68a
Eurofit	1230.7a	7.08a	42.11ab	70.55a
Akteur	1772.4b	6.74b	41.55a	69.17a
Meritto	1491.8ab	7.15a	44.15b	74.68b

Values in the same column, with different letters are statistically significant at $p \le 0.05$.

TKW - thousand kernels weight; VW - volume weight

It is known, that FHB can lead to the serious yield losses, due to production of small-sized grains and formation of some infertile spikelets (Eggert et al. 2010).

In our case, grain was sieved after the harvest, using the sieves 2.2 x 22 mm to remove the smallest, waste wheat grains; then TKW (thousand kernels weight) and VW (volume weight) were determined. Regardless this sieving, significant difference in VW and in TKW between the inoculated variants and the check were found (Table 3).

Our results concerning the grain yield evaluation (Table 3) showed statistically

significant differences between individual experimental variants in dependence on way of treatment – especially variants with artificial inoculation differed significantly from variants with fungicidal treatment and from the check variant. Protective fungicidal treatment by Prosaro proved positive effect again – significantly increasing of yield in variants combining artificial inoculation and fungicidal treatment (IV,VI, VIII) compared to inoculated variants without fungicidal treatment (III, V, VII) was found.

Statisticaly significant differences were determined also between the individual years

and individual evaluated cultivars – cultivar Meritto from quality group B exceeded in yield cultivar Eurofit from quality group A and especially cultivar Akteur from quality group E.

Baking quality parameters

Wang et al. (2005) concluded on the basis of their investigation that the crude

protein content in the wheat grain was not affected by the infection with *F. culmorum*. In comparison, an increase in protein content in infected grain was reported by Boyacioglu and Hettiarachchy (1995) and slight decrease in protein content by Dexter et al. (1997). In our case, effect of *Fusarium* spp. infection grade on protein and wet gluten content in grain dry matter was not significant (Table 4).

Table 4. Average values of quality parameters for individual variants, years and wheat cultivars

Spacification	СР	WG	ZS	FN	FY	AC	LV	H/D
Specification	(%)	(%)	(ml)	(s)	(%)	(%)	(ml)	
Ι	14.82a	31.11a	52.6a	308.2b	66.2a	0.58a	269.4d	0.65d
II	14.65a	31.52a	53.0a	299.2b	65.9ab	0.58a	266.3cd	0.61cd
III	14.64a	31.94a	43.0bc	219.4ac	65.1ab	0.69b	236.2a	0.46a
IV	14.78a	31.95a	44.9b	239.6a	64.6ab	0.67b	243.9ab	0.55bc
V	14.61a	31.72a	42.9bc	232.3a	63.7bc	0.69b	244.0ab	0.54bc
VI	14.42a	31.11a	46.2b	252.0a	65.4ab	0,59a	255.3bc	0.60cd
VII	14.66a	32.00a	40.6c	194.0c	62.6c	0.69b	232.5a	0.44a
VIII	14.51a	31.54a	44.1b	215.1c	64.4ab	0.67b	246.0ab	0.53ab
2008	14.98a	31.52ab	53.2a	297.2b	62.5a	0.67b	249.1b	0.52a
2009	14.75a	32.22b	45.5b	326.6c	68.1c	0.63a	257.1c	0.61b
2010	14.20b	31.11a	39.0c	111.2a	63.6b	0.63a	241.3a	0.52a
Eurofit	14.85a	32.25a	47.5a	240.0a	65.6b	0.64a	253.0b	0.55a
Akteur	15.28b	32.96a	48.1a	258.8b	66.7c	0.64a	251.9b	0.55a
Meritto	13.80c	29.61b	42.1b	236.3a	61.9a	0.65a	242.6a	0.54a

Values in the same column with different letters are statistically significant at $p \le 0.05$.

CP – crude protein content in grain dry matter; WG – wet gluten content in grain dry matter; ZS – Zeleny. sedimentation; FN – falling number; FY – flour yield; AC – ash content of flour; LV – loaf volume of bread; H/D – rate of height and diameter of bread.

The Zeleny sedimentation index measures the swelling potential of the kernel protein. Gärtner et al. (2008) observed general reduction of Zeleny index in wheat grains after the FHB infection. This indicates that although the total amount of protein remains quite stable, the infection may alter its quality. According to Hareland (2003), fungal expected increase the infection is to degradation of starch, due to the presence and activity of enzymes such as α -amylase in the kernels, which is measurable by means of the falling number. These findings are in accordance with our results - ZS and FN showed distinctively decreased values in the inoculated variants (the worst for variant VII artificial inoculation at the beginning and at the end of wheat flowering). Our results also showed significant differences in ZS and FN between the individual years - low values of these parameters in the year 2010 were caused by very rainy conditions during the grain maturing and harvest (Table 4).

A slight increase in ash content of flour was recorded in the artificially inoculated variants. Ash is mainly composed of minerals of the seed coat (bran and aleurone). The proportion of ash in flour is therefore an indicator of its purity, as increasing ash content indicates an alteration in the kernel seed coat - volume ratio. It is possible to associate this observation with the presence of shrivelled. misshapen fusarium-infected kernels (Jones, Mirocha, 1999). Lower values of the flour yield in more infected variants were confirmed by our results. But, it is also evident from the results, that the flour yield was more affected by years than by experimental variants. The loaf volumes of bread and ratios between the height and diameter of bread had a descending tendency with progressive infection and DON content.

These results are in agreement with those of Dexter et al. (1996), who mentioned the decrease of loaf volume of wheat bread following *Fusarium* spp. infection.

Mixolab characteristics

The average values of Mixolab characteristics for specific variants, years and wheat varieties are shown in Table 5. Point C1 was not included in the final evaluation because this side point is sufficiently

represented by the difference between points C1 and C2 (C1-C2).

Mixolab detected sensitively the effect of the intensity of *Fusarium* spp. infection on rheological properties. The value of dough torque for parameter C2, which represents the dough strength, decreased in inoculated variants (especially in variants III and VII). This is in accordance with the values of other technological parameters related to protein, like Zeleny sedimentation index.

Table 5. Average values of Mixolab characteristics for individual variants, years and wheat cultivars

Specification	C2	C3	C4	C5	C1-C2	C3-C4	C5-C4	DS
Specification	(nm)	(min)						
Ι	0.40ab	1.70ab	1.35b	1.77a	0.69a	0.36a	0.42a	7.3b
II	0.37ab	1.74b	1.33b	1.70a	0.72ab	0.41a	0.37a	6.7ab
III	0.30bc	1.60ab	1.16ab	1.39b	0.80b	0.44a	0.23b	5.3a
IV	0.32bc	1.62ab	1.20ab	1.51ab	0.76ab	0.43a	0.32ab	5.5a
V	0.31bc	1.62ab	1.18ab	1.61ab	0.76ab	0.44a	0.42a	5.9ab
VI	0.45a	1.66ab	1.23ab	1.66a	0.67ab	0.38a	0.44a	6.8ab
VII	0.25c	1.50a	1.02a	1.35b	0.82b	0.48a	0.32ab	5.3a
VIII	0.38ab	1.49a	1.10ab	1.52ab	0.77ab	0.40a	0.63c	5.3a
2008	0.37a	1.81a	1.45a	1.97a	0.74a	0.36a	0.52a	6.5a
2009	0.44b	2.05b	1.74b	2.39b	0.65b	0.31a	0.65a	7.5b
2010	0.24c	0.99c	0.40c	0.33c	0.86c	0.58b	0.20b	4.0c
Eurofit	0.35ab	1.73a	1.29a	1.64a	0.74ab	0.44ab	0.35a	6.6a
Akteur	0.40b	1.61b	1.28a	1.83a	0.72a	0.32a	0.83b	6.9a
Meritto	0.30a	1.51c	1.02b	1.22b	0.78b	0.49b	0.20a	4.5b

Values in the same column with different letters are statistically significant at $p \le 0.05$

C2 – protein weakening; C3 – starch gelatinization; C4 – stability of gel; C5 – starch retrogradation; C1-C2 – fall of protein strenght; C3-C4 – diastatic activity; C5-C4 – anti-stalling effect; DS – time of dough stability before weakening.

As was predicted from the values of falling number, Mixolab characteristics C3 and C4, which imply the starch damage, were again the worst for artificially inoculated variants III and VII. The dough with such characteristics is usually stickier and can have a poor baking quality. The low value of Mixolab characteristic C5 for these variants, which represents the rate of retrogradation, verifies the worse quality of the starch part of the wheat grain (Collar et al., 2006). The final bread from such dough may have a similar value of the loaf volume, but it is possible to anticipate undesirable changes of the shape (Nightingale et al., 1999).

Mixolab characteristic C1-C2, which represents the protein network strength under increasing heating, as well as characteristic C3-C4 which represents diastatic activity, show the highest values, i.e. the highest decrease, in variants III and VII with the highest infection grade.

On the other hand, variants I and II with the lowest intensity of infection have the best stability in the protein part of curve. In the starch part the highest increase of viscosity was observed in these variants (I and II), which indicated undamaged starch grains. Mixolab characteristic C5-C4, which represents the shelf life of the end product, reaches the worst, lowest values for variants III and VII and the best values for variants I and II.

Mixolab also distinguished sensitively the variations of baking quality among the cultivars and also the shifts in the characteristics in the individual years. The higher quality cultivars Akteur and Eurofit overtopped the worse quality cultivar Meritto in all evaluated characteristics.

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The significant correlations found between the Mixolab characteristics and standard technological parameters, especially ZS and FN, confirmed the already published knowledge (Ozturk et al., 2008; Codina et al., 2010). Significant correlations were also found between Mixolab characteristics and flour yield, as well as between Mixolab characteristics and baking parameters (LV; ratio of H/D). In agreement with our expectations, the higher values of Mixolab characteristics C2 to C5 showed positive correlations with the baking characteristics, whereas the increased differences C1-C2 and C3-C4 indicated deterioration of the breadmaking quality (Table 6).

	СР	WG	ZS	FN	LV	H/D	FY	AC
C2	0.24*	0.19	0.45**	0.54**	0.50**	0.44**	0.43**	-0.21
C3	0.45**	0.30^{*}	0.61**	0.91**	0.49**	0.37**	0.43**	-0.03
C4	0.49**	0.34**	0.64**	0.91**	0.53**	0.39**	0.45**	-0.03
C5	0.55**	0.41**	0.64^{**}	0.91**	0.53**	0.38**	0.47**	-0.01
C1C2	-0.29*	-0.19	-0.53**	-0.76**	-0.62**	-0.59**	-0.55**	0.29*
C3C4	-0.39**	-0.30*	-0.44**	-0.55**	-0.43**	-0.29*	-0.34**	0.05
C5C4	0.24	0.26*	0.24^{*}	0.25*	0.25^{*}	0.19	0.29*	-0.08
DS	0.66**	0.47^{**}	0.69**	0.78^{**}	0.64**	0.48^{**}	0.60^{**}	-0.25*
DON	0.06	0.06	-0.27*	-0.40**	-0.71**	-0.85**	-0.48**	0.74^{**}

Table 6. Correlations among the evaluated parameters

CONCLUSIONS

Our results confirmed that FHB is able to cause not only significant yield reduction, but also leads to the occurrence of numerous mycotoxins in grain. The most common Fusarium toxin produced on winter wheat, DON, is presented in this work. Increasing intensity of *Fusarium* spp. infestation worsened the rheological quality and led to the decrease of most of the evaluated baking parameters, especially Zeleny sedimentation index, falling number and the main baking criterion - loaf volume of bread. Besides different indirect agricultural measures and choosing of the more resistant cultivars, a direct measure consisting in protective chemical treatment may contribute to the reduction in the risk of FHB disease and toxin contamination.

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