

## Changes in Baking Quality of Winter Wheat with Different Intensity of *Fusarium* spp. Contamination Detected by Means of New Rheological System Mixolab

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### Abstract

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The aim of our work was to assess the possibility of detecting the changes in the baking quality of winter wheat with different levels of *Fusarium* spp. contamination using a new rheological system Mixolab, and to determine the correlations between the Mixolab characteristics and other quality parameters of wheat flour and grain. The standard technological characteristics (crude protein, Zeleny sedimentation index, wet gluten, falling number), loaf volume, shape features of bread (height and diameter), Mixolab parameters, and mycotoxin deoxynivalenol (DON) content were determined in 3 winter wheat cultivars (Akteur – quality group E – elite; Eurofit – quality group A; Meritto – quality group B) with different levels of *Fusarium* spp. contamination (8 variants) in two years. Increasing intensity of *Fusarium* spp. contamination evidently worsened the rheological quality and its negative effects on protein and mainly on the starch part of the grain was obvious in Mixolab curves. High correlations were found between Mixolab characteristics and standard technological parameters, as well as between Mixolab parameters and the main baking criterion – loaf volume.

**Keywords:** winter wheat; *Fusarium* spp.; bread making quality; Mixolab

Mixolab is a relatively new system, accepted as the ICC standard method No. 173 (ICC 2006), which makes possible to evaluate physical dough properties such as dough stability or weakening, and starch characteristics in one measurement

(KAHRAMAN *et al.* 2008). It is enabled by excessive mixing and controlled heating of the kneader to 90°C and subsequent cooling to 50°C. Mixolab records in real time the torque (in nm) produced by the dough between two blades. A typical Mixolab

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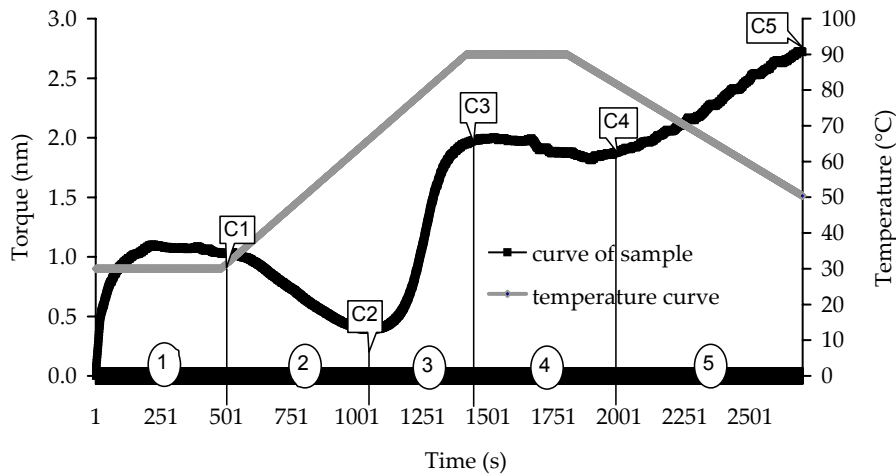


Figure 1. Standard Mixolab curve

curve is separated into five stages, characterised by 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 hot-formed gel
- C5 represents starch retrogradation during the cooling period
- C1–C2 indicates the protein network strength under increasing heating
- C3–C4 shows diastatic activity and relates with falling number
- C5–C5 correlates with the anti-staling effects, represents the shelf life of the end products

Dough stability indicates the stability of the dough before weakening

The two first stages of the curve correspond to the rheological characteristics of stability, elasticity, and water absorption, related to proteins. The consistency of dough, which decreases with excessive mixing, 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 activity. The increase in the consistency curve is mainly due to starch gelatinisation. At the fourth stage, consistency decreases as a result of amylolytic activity 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, and it is also related to the starch retrogradation (ROSELL *et al.* 2007). This stage depends mainly on the level of amylose present, which starts the process of retrogradation. For example, for waxy wheat that contains only a low level of amylose, no increase of the curve is detected (GRAYBOSCH *et al.* 2003).

Mixolab parameters show, based on the results published up to now, high compatibility with standard rheological analysis (for example farinograph, extensograph, or amylograph). Consequently, it is possible to anticipate a potential prediction of bread making quality of common wheat from these parameters (CATO & MILLS 2008). But there are hardly any studies on the efficiency of this Mixolab system to predict rheological parameters of wheat with changed characteristics caused by fungi species, which have become a serious problem in the wheat cultivation during recent years. This fungi contamination, in cereals mainly *Fusarium* spp. contamination, causes not only grain yield loss but, because of their secondary metabolites mycotoxins, has also a negative health impact (GARCIA *et al.* 2009). The most important is the trichothecene mycotoxin deoxynivalenol (DON), considered as a contamination marker subjected to the European Commission Regulation (EC 2006). DON maximum level was set at 1250 µg/kg for unprocessed cereals and 750 µg/kg for cereal flours.

Besides these facts, several authors also mentioned negative effects of *Fusarium* infection on bread making quality of wheat and the reduction of loaf volumes (SEITZ *et al.* 1986; NIGHTINGALE *et al.* 1999; GÄRTNER *et al.* 2008). On the other hand, there are some contradictory studies where

a strong *Fusarium* contamination did not significantly influence the bread making properties (ANTES *et al.* 2001; PRANGE *et al.* 2005).

The aim of this work is (i) to confirm the correlations between the Mixolab characteristics and other quality parameters of wheat flour and grain; (ii) to detect the changes in the baking quality of winter wheat due to different levels of *Fusarium* spp. contamination using the new rheological system Mixolab.

## MATERIAL AND METHODS

**Plant material.** A set of 24 winter wheat samples from the exact field plot trial, conducted in the years of 2008 and 2009 on the experimental station of the Department of Crop Production of the Czech University of Life Sciences Prague in Uhřetěves (295 m above sea level, average annual temperature 8.4°C, average sum of precipitation 575 mm), was used for the evaluation of the bread making quality and DON content in grain. The set included 3 winter wheat cultivars (Akteur – quality group E; Eurofit – quality group A; Meritto – quality group B) and 8 variants coming from the field plot trial.

**Artificial inoculation and evaluation of *Fusarium* spp. infestation.** Different conditions were used of artificial inoculation of wheat with *Fusarium* spp. and simultaneously several ways of fungicide treatment, with the aim to obtain a scale of samples with different levels of *Fusarium* spp. contamination. The list of variants is given in Table 1.

Table 1. List of the field plot variants

Variant signification	Treatment
I	preventive fungicidal treatment
II	check variant without any treatment
III	artificial inoculation at the end of flowering + fungicide
IV	artificial inoculation at the end of flowering
V	artificial inoculation at the beginning of flowering + fungicide
VI	artificial inoculation at the beginning and at the end of flowering + fungicide
VII	artificial inoculation at the beginning of wheat flowering
VIII	artificial inoculation at the beginning and at the end of flowering

The isolates of *F. culmorum* and *F. graminearum* used for the artificial inoculation were obtained from the mycological collection of the Crop Research Institute in Prague and cultivated on sterile wheat grains. The preparation of inoculums for the application: 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,  $10^7$  of spores/ml (Bürker chamber was used for the verification of inoculums density), 2 l of suspension per experimental plot (12 m<sup>2</sup>). 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 (effective substances prothioconazole and tebuconazole) was applied two days before the inoculation.

Grain samples were taken for the evaluation after the wheat harvest. PCR assay was used for *Fusarium* spp. detection. Total DNA was extracted from the grain samples using the DNeasy Plant Mini Kit (Qiagen, Hilden, Germany). The sequences of the primers, PCR reaction mixture, and the conditions used for detecting *F. culmorum* and *F. graminearum* in this study were identical to those described by SCHILLING *et al.* (1996). This type of PCR assay enables to distinguish only the intensity of *Fusarium* spp. attack without quantitative formulation, therefore the DON content was considered as an indicator of the intensity of *Fusarium* spp. contamination.

**Deoxynivalenol content.** UPLC-TOF MS system consisting of Ultra-Performance Liquid Chromatography (Acquity, Waters, USA) coupled to a time-of-flight mass spectrometer (LCT Premier, Waters, USA) was used for the determination of deoxynivalenol (DON) in wheat grain. All of wheat samples were milled and homogenised prior to the extraction procedure. The representative sample (12.5 g) was extracted with 50 ml of acetonitrile: water mixture (84:16, v/v) for 60 minutes. An aliquot of the 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. For the separation of analytes, Acquity UPLC HSS T3 column (100 × 2.1 mm *i.d.*, 1.7 µm particle size, Waters, USA) was used. The working parameters of the analytical method were established as follows: flow rate 0.3 µl/min, column temperature 40°C,

injection volume 5 µl, auto-sampler temperature 10°C, the mobile phase consisted of 5mM ammonium formiate in water (A) and methanol (B). The fast linear gradient program for the separation of the target compounds was used. The orthogonal time-of-flight mass spectrometer was operated in the negative electro-spray ionisation (ESI-) mode. Capillary voltage was set at 3500 V, cone voltage 40 V, source temperature 120°C, desolvation temperature 350°C. Nitrogen was used as a solvent as well as cone gas; its flow rates were 750 l/h and 10 l/h, respectively. Target analytes were identified according to their retention times and accurate masses. Software MassLynx 4.1 with the application of QuanLynx manager was used for the data acquisition and processing.

**Sample preparation for technological quality evaluation.** The wheat grain samples were purified using sieves (2.2 × 22 mm) and conditioned to 15.5% moisture before milling on a Bühler mill automat MLU 202. Milling proceeded in two sections divided into three parts with different conditions of grinding and sieving. 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.

**Bread making quality analyses.** Within the frame of the bread making quality, crude protein content (CP) according to Kjeldahl method (EN ISO 20483; ICC-Standard No.105/2), 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, and ash content (AC) – ISO 2171 were determined.

Bread was made according to the internal protocol from flour (300 g), yeast (12 g), fat (3 g), sugar (4.5 g), 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 ball-shaped parts and left in a thermostat for 50 minutes. Afterwards, the pieces were put in the oven at 240°C for 14 minutes. 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.

**Mixolab determination.** Rheological characteristics were determined using the apparatus Mixolab

(Chopin, Tripette et Renaud, Paris, France) according to the Mixolab protocol Chopin+ for white flour (Mixolab appl. Handbook, 2008). Point C1 was not included in the final evaluation because this side point is sufficiently represented by the difference between points C1 and C2 (C1C2).

**Statistical analysis.** The results were statistically evaluated by the one-way 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 in the software STATISTICA 8.0 CZ (StatSoft).

## RESULTS AND DISCUSSION

### Intensity of *Fusarium* spp. infection and DON content

The levels of *Fusarium* spp. infection and DON content in the individual grain samples are summarised in Table 2, where the differences are visible between the individual years, experimental variant, and cultivars. In 2008, the infection grade was evidently higher than in 2009. It was most likely due to the climatic conditions during wheat flowering, which were favourable for a fast development of *Fusarium* inoculum in that year.

There are conflicting reports on the correlation between the *Fusarium* infection grade and DON content. Some authors did not confirm positive correlation between the infection grade and DON content (KUSHIRO 2008; GARCIA *et al.* 2009) while others found a high positive significant correlation (SNIJDERS 2004). Nevertheless, in the case of a strong infection pressure evoked by artificial inoculation is it possible to suppose also a high DON content.

In our study, we found the highest average content of deoxynivalenol (DON) in grain for variants VII and VIII with the highest intensity of infection (Table 3). These detected average values of DON (3023, resp. 4003 µg/kg) exceeded considerably the limit permitted for the food grain, nevertheless, so intensive infection enables a very good distinction of the effect of *Fusarium* spp. infestation on the wheat characteristics. Significant differences were observed between DON contents of the individual variants (I–III and VII–VIII) as well as between the two monitored years but not between the cultivars.

Table 2. *Fusarium* infection grade of wheat samples (PCR assay using specific primers) and individual DON content in grain

Variant	Cultivar	2008			2009		
		<i>F. culmorum</i>	<i>F. graminearum</i>	DON content (µg/kg)	<i>F. culmorum</i>	<i>F. graminearum</i>	DON content (µg/kg)
I	Meritto	+	–	n.d.	+	–	148
	Akteur	+	–	12	+	–	148
	Eurofit	–	–	n.d.	+	–	54
II	Meritto	++	+	266	+	+	111
	Akteur	++	–	151	+	–	117
	Eurofit	++	–	211	–	+	4
III	Meritto	++	–	337	+	–	117
	Akteur	++	+	756	+	–	174
	Eurofit	++	–	324	–	–	502
IV	Meritto	+	–	638	+	–	436
	Akteur	+++	+	1695	++	–	2148
	Eurofit	+	–	578	+	–	271
V	Meritto	++	–	2324	+	–	1114
	Akteur	++	–	2433	+	–	764
	Eurofit	++	–	1489	+	–	994
VI	Meritto	++	+	2443	+	–	727
	Akteur	++	–	3556	+	–	1418
	Eurofit	++	+	2388	+	–	294
VII	Meritto	+++	+	6146	++	+	664
	Akteur	+++	+	6304	++	+	632
	Eurofit	++	+	3889	++	+	1053
VIII	Meritto	+++	+	7335	++	+	1700
	Akteur	+++	+	5747	++	+	1568
	Eurofit	++	–	6595	++	–	1053

+ sample positive (low infection grade); ++ sample positive (medium infection grade); +++ sample positive (high infection grade); – sample negative; LOQ (DON) = 5 µg/kg; n.d. – non detected

### Effect of *Fusarium* spp. infection on Mixolab parameters

The average values of Mixolab characteristics for specific variants, years, and wheat cultivars and their statistical comparisons are shown in Table 3. Visible variations were found between the variants, years, as well as cultivars.

The value of dough torque for parameter C2 represents the dough strength decreases from variants I and II to the most infected variant VIII. It is in accordance with the values of the technological characteristics relating to protein like wet gluten and Zeleny sedimentation index.

As was predicted from the values of the falling number, Mixolab parameters C3 and C4 which imply the starch damage were again the worst for variant VIII. The dough with such results is usually stickier and can have a poor baking quality (DEXTER *et al.* 1985). In our case, the low value of Mixolab characteristic C5 for this variant, 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), as confirmed by our results (Figure 2).

Table 3. Average values of Mixolab characteristics and DON content for individual variants, years and wheat cultivars

	C2 (nm)	C3 (nm)	C4 (nm)	C5 (nm)	C1C2 (nm)	C3C4 (nm)	C5C4 (nm)	DS (min)	DON ( $\mu\text{g}/\text{kg}$ )
I	0.46 <sup>cd</sup>	2.04 <sup>c</sup>	1.76 <sup>bc</sup>	2.44 <sup>bc</sup>	0.66 <sup>ab</sup>	0.29 <sup>a</sup>	0.68 <sup>bc</sup>	7.8 <sup>ab</sup>	40.7 <sup>a</sup>
II	0.47 <sup>d</sup>	2.06 <sup>c</sup>	1.80 <sup>c</sup>	2.55 <sup>c</sup>	0.63 <sup>b</sup>	0.26 <sup>a</sup>	0.76 <sup>c</sup>	8.2 <sup>b</sup>	162.9 <sup>a</sup>
III	0.44 <sup>bcd</sup>	2.00 <sup>bc</sup>	1.65 <sup>abc</sup>	2.28 <sup>abc</sup>	0.66 <sup>ab</sup>	0.35 <sup>a</sup>	0.63 <sup>abc</sup>	7.6 <sup>ab</sup>	312.2 <sup>a</sup>
IV	0.40 <sup>abc</sup>	1.93 <sup>abc</sup>	1.62 <sup>abc</sup>	2.27 <sup>abc</sup>	0.69 <sup>abc</sup>	0.31 <sup>a</sup>	0.64 <sup>abc</sup>	7.2 <sup>ab</sup>	961.0 <sup>ab</sup>
V	0.39 <sup>abc</sup>	1.90 <sup>ab</sup>	1.53 <sup>abc</sup>	2.13 <sup>ab</sup>	0.71 <sup>abc</sup>	0.36 <sup>a</sup>	0.6 <sup>abc</sup>	6.3 <sup>a</sup>	1519.7 <sup>ab</sup>
VI	0.37 <sup>ab</sup>	1.84 <sup>a</sup>	1.47 <sup>a</sup>	1.94 <sup>a</sup>	0.73 <sup>bc</sup>	0.37 <sup>a</sup>	0.48 <sup>ab</sup>	6.7 <sup>a</sup>	1804.3 <sup>abc</sup>
VII	0.36 <sup>a</sup>	1.86 <sup>ab</sup>	1.51 <sup>ab</sup>	1.93 <sup>a</sup>	0.74 <sup>bc</sup>	0.35 <sup>a</sup>	0.42 <sup>a</sup>	6.1 <sup>a</sup>	3022.8 <sup>bc</sup>
VIII	0.35 <sup>a</sup>	1.81 <sup>a</sup>	1.43 <sup>a</sup>	1.91 <sup>a</sup>	0.75 <sup>c</sup>	0.38 <sup>a</sup>	0.49 <sup>ab</sup>	6.3 <sup>a</sup>	4002.7 <sup>c</sup>
2008	0.37 <sup>a</sup>	1.81 <sup>a</sup>	1.45 <sup>a</sup>	1.97 <sup>a</sup>	0.74 <sup>b</sup>	0.36 <sup>a</sup>	0.52 <sup>a</sup>	6.5 <sup>a</sup>	2317.4 <sup>b</sup>
2009	0.44 <sup>b</sup>	2.05 <sup>b</sup>	1.74 <sup>b</sup>	2.39 <sup>b</sup>	0.65 <sup>a</sup>	0.31 <sup>a</sup>	0.65 <sup>b</sup>	7.5 <sup>b</sup>	639.2 <sup>a</sup>
EUROFIT	0.43 <sup>a</sup>	2.03 <sup>a</sup>	1.75 <sup>a</sup>	2.31 <sup>b</sup>	0.68 <sup>a</sup>	0.28 <sup>a</sup>	0.57 <sup>b</sup>	7.8 <sup>a</sup>	1175.7 <sup>a</sup>
AKTEUR	0.42 <sup>a</sup>	2.01 <sup>a</sup>	1.73 <sup>a</sup>	2.54 <sup>c</sup>	0.68 <sup>a</sup>	0.28 <sup>a</sup>	0.81 <sup>c</sup>	8.4 <sup>a</sup>	1727.6 <sup>a</sup>
MERITTO	0.37 <sup>b</sup>	1.75 <sup>b</sup>	1.31 <sup>b</sup>	1.69 <sup>a</sup>	0.72 <sup>b</sup>	0.44 <sup>b</sup>	0.38 <sup>a</sup>	5.0 <sup>b</sup>	1531.6 <sup>a</sup>

Values with different letter combinations are statistically significant at  $P \leq 0.05$  (probability of error max. 5%); LOQ (DON) = 5  $\mu\text{g}/\text{kg}$ ; C2 – protein weakening; C3 – starch gelatinization; C4–stability of gel; C5 – starch retrogradation; C1C2 – fall of protein strength; C3C4 – diastatic activity; C5C4 – anti-stalling effect; DS – stability of the dough before weakening

Mixolab distinguished sensitively the variations among the baking quality of the cultivars and also the shifts in the characteristics in the individual years (Figure 3). According to the figure the cultivars kept their features in compliance with their quality group. The higher quality cultivars, Akteur and Eurofit, overtopped the worse quality cultivar Meritto in both years monitored. In 2008, when *Fusarium* spp. infection was higher, there was a clear shift in the curves to a worse baking quality than in 2009. Higher differences were recorded for the starch part of the wheat grain, which could have been caused by an early hydrolysis of damaged starch in more infected grains (BARRERA *et al.* 2007).

The sensitivity of Mixolab detection of single variants with a different intensity of *Fusarium* spp.

contamination is demonstrated on the cultivar Eurofit (Figure 4). Clearly shown is the effect of the intensity of *Fusarium* spp. infection on rheological characteristics and the shift of individual curves. Variants I and II with the lowest intensity of infection displayed the best stability and the lowest decline of the curve in the protein part. In the starch part the highest increase of viscosity was observed which indicated undamaged starch grains. On the contrary, variants VII and VIII showed a considerable fall of dough consistency, which denotes inferior quality of gluten, and a worse course of the curve in the starch part. This result is in agreement with the low falling numbers of these variants. The last part of the curve also showed the smallest increase as a result of the low viscosity of the dough (GRANT 1998).



Figure 2. Treated variant I and infected variant VII – cultivar Akteur (in 2008)

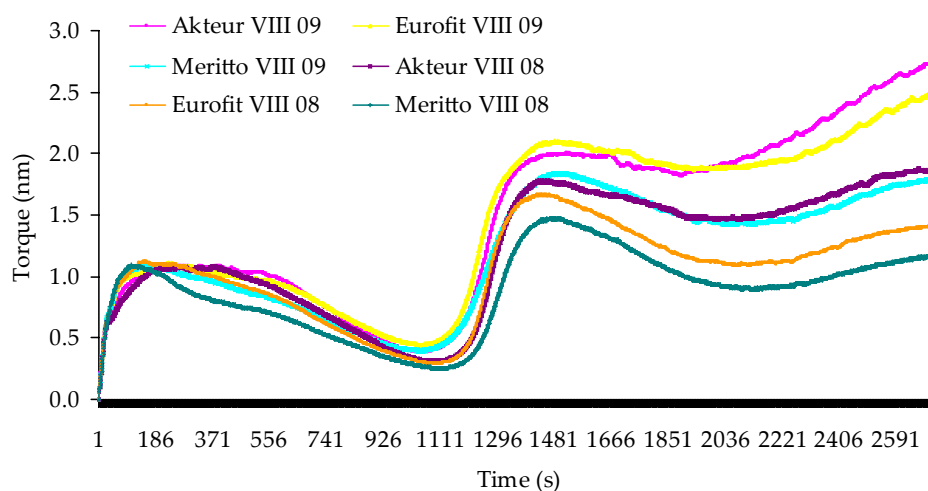


Figure 3. Mixolab curves for individual cultivars in 2008 and 2009 (variant VIII)

### Changes in technological and baking characteristics

The effect of *Fusarium* spp. infection was visible in both protein and starch parts of the wheat grain and markedly influenced the technological characteristics of the grain (Table 4). Some authors mentioned an increase of protein content after the *Fusarium* spp. contamination (BOYACIOĞLU & HETTIARACHCHY 1995), but others, for example DEXTER *et al.* (1997) and GÄRTNER *et al.* (2008), mentioned a slight decrease of it. In our case, slight decrease occurred of protein and wet gluten contents in the most infected variant VIII in comparison with variants I and II. The Zeleny sedimentation index measures the swelling potential of the kernel protein. MEYER *et al.* (1986) and GÄRTNER *et al.* (2008) observed general reduction of Zeleny index in wheat grains after 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 infection is expected to increase the degradation of starch due to the presence of enzymes such as  $\alpha$ -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 values for variant VIII). An increase of 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

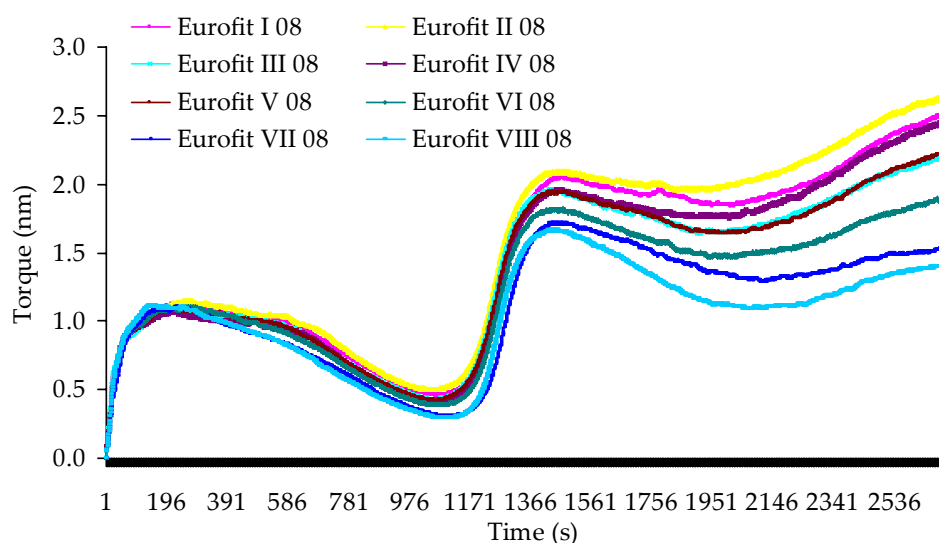


Figure 4. Mixolab curves for cultivar Eurofit in 2008 (variants I–VIII)

Table 4. Average values of quality characteristics for individual variants (I-the lowest; VIII-the highest grade of DON content), years and wheat cultivars

	CP (%)	WG (%)	ZS (ml)	FN (s)	LV (ml)	H/D	FY (%)	AC (%)
I	15.0 <sup>ab</sup>	32.2 <sup>ab</sup>	57.0 <sup>b</sup>	365.3 <sup>cd</sup>	272.9 <sup>c</sup>	0.62 <sup>bc</sup>	65.79 <sup>ab</sup>	0.59 <sup>b</sup>
II	15.3 <sup>b</sup>	33.2 <sup>b</sup>	56.7 <sup>b</sup>	380.0 <sup>d</sup>	275.5 <sup>c</sup>	0.66 <sup>c</sup>	65.57 <sup>ab</sup>	0.59 <sup>b</sup>
III	14.7 <sup>ab</sup>	31.7 <sup>ab</sup>	49.7 <sup>a</sup>	330.2 <sup>bcd</sup>	259.9 <sup>bc</sup>	0.60 <sup>bc</sup>	66.66 <sup>b</sup>	0.60 <sup>b</sup>
IV	14.6 <sup>ab</sup>	31.5 <sup>ab</sup>	46.8 <sup>a</sup>	303.0 <sup>abc</sup>	250.2 <sup>ab</sup>	0.60 <sup>bc</sup>	65.62 <sup>ab</sup>	0.68 <sup>a</sup>
V	14.8 <sup>ab</sup>	31.9 <sup>ab</sup>	47.7 <sup>a</sup>	298.2 <sup>ab</sup>	246.1 <sup>ab</sup>	0.57 <sup>abc</sup>	65.01 <sup>ab</sup>	0.69 <sup>a</sup>
VI	14.6 <sup>ab</sup>	31.5 <sup>ab</sup>	47.7 <sup>a</sup>	290.3 <sup>ab</sup>	246.8 <sup>ab</sup>	0.54 <sup>abc</sup>	65.56 <sup>ab</sup>	0.69 <sup>a</sup>
VII	14.7 <sup>ab</sup>	31.7 <sup>ab</sup>	45.2 <sup>a</sup>	270.5 <sup>ab</sup>	237.8 <sup>a</sup>	0.49 <sup>ab</sup>	63.51 <sup>a</sup>	0.70 <sup>a</sup>
VIII	14.5 <sup>a</sup>	31.2 <sup>a</sup>	44.2 <sup>a</sup>	257.7 <sup>ab</sup>	235.7 <sup>a</sup>	0.45 <sup>a</sup>	64.94 <sup>ab</sup>	0.71 <sup>a</sup>
2008	14.9 <sup>b</sup>	31.5 <sup>a</sup>	53.2 <sup>b</sup>	297.2 <sup>a</sup>	249.1 <sup>a</sup>	0.52 <sup>a</sup>	62.54 <sup>a</sup>	0.64 <sup>b</sup>
2009	14.7 <sup>a</sup>	32.2 <sup>b</sup>	45.5 <sup>a</sup>	326.6 <sup>b</sup>	257.1 <sup>b</sup>	0.61 <sup>b</sup>	68.12 <sup>b</sup>	0.67 <sup>a</sup>
EUROFIT	15.1 <sup>b</sup>	32.8 <sup>a</sup>	51.1 <sup>a</sup>	305.1 <sup>a</sup>	258.8 <sup>a</sup>	0.57 <sup>a</sup>	66.17 <sup>b</sup>	0.65 <sup>a</sup>
AKTEUR	15.6 <sup>c</sup>	33.5 <sup>a</sup>	52.2 <sup>a</sup>	332.4 <sup>a</sup>	255.1 <sup>a</sup>	0.57 <sup>a</sup>	67.47 <sup>b</sup>	0.66 <sup>a</sup>
MERITTO	13.7 <sup>a</sup>	29.3 <sup>b</sup>	44.8 <sup>b</sup>	298.2 <sup>b</sup>	245.5 <sup>b</sup>	0.56 <sup>a</sup>	62.36 <sup>a</sup>	0.66 <sup>a</sup>

Values with different letter combinations are statistically significant at  $P \leq 0.05$  (probability of error max. 5%); CP – crude protein; WG – wet gluten; ZS – Zeleny sedimentation; FN - falling number; LV – loaf volume; H/D – rate of height and diameter of bread; FY – flour yield; AC – ash content of flour

(JONES & MIROCHA 1999). This observation is probably in connection with the lower values of the flour yield with more infected variants which was also confirmed by our results. Likewise, the loaf volumes 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 reported clear deterioration of bread making quality owing to high *Fusarium* spp. contamination.

#### Relationship between Mixolab parameters, DON content, technological and baking characteristics

Highly significant negative correlations were found between the values of Mixolab individual points (C2, C3, C4, C5) and DON content indicating the intensity of *Fusarium* spp. contamination. This shows a high response of Mixolab to the changes of wheat grain caused by *Fusarium* spp. infection (Table 5).

The significant relations found between Mixolab characteristics and standard technological parameters, especially ZS and FN, confirmed the already

published knowledge (OZTURK *et al.* 2008; CODINĂ *et al.* 2010). Highly significant correlations were also found between Mixolab and flour characteristics (flour yield; ash content) as well as between Mixolab and baking parameters (LV; ratio of H/D). According to our presumption the higher values of Mixolab parameters C2 to C5 showed positive relations with the baking characteristics whereas the increasing differences C1C2 and C3C4 indicated deterioration of the bread making quality.

#### CONCLUSION

The high sensitivity of Mixolab system for monitoring the changes in rheological characteristics of winter wheat with different intensities of *Fusarium* spp. contamination was confirmed in our study. Increasing intensity of *Fusarium* spp. contamination worsened rheological quality and hence took up a negative effect on protein and mainly on the starch part of Mixolab curves. The evaluated wheat cultivars kept the bread making quality according to their quality classification notwithstanding the infection grade of the individual cultivars.

High correlations were found between Mixolab characteristics and the standard technological param-



Table 5. Correlations among the evaluated parameters

	CP	WG	ZS	FN	LV	H/D	FY	AC
C2	0.37**	0.53**	0.35**	0.80**	0.75**	0.77**	0.68**	-0.72**
C3	0.51**	0.68**	0.23	0.64**	0.64**	0.66**	0.83**	-0.56**
C4	0.58**	0.71**	0.30*	0.58**	0.62**	0.59**	0.74**	-0.50**
C5	0.65**	0.75**	0.36*	0.69**	0.62**	0.61**	0.80**	-0.54**
C1C2	-0.27	-0.45**	-0.18	-0.75**	-0.65**	-0.75**	-0.69**	0.66**
C3C4	-0.54**	-0.59**	-0.34*	-0.36*	-0.46**	-0.36*	-0.44**	0.28*
C5C4	0.62**	0.66**	0.37**	0.69**	0.50**	0.51**	0.72**	-0.48**
DS	0.75**	0.81**	0.47**	0.59**	0.59**	0.48**	0.68**	-0.50**
DON	-0.04	-0.19	-0.18	-0.77**	-0.72**	-0.85**	-0.51**	0.71**

\*correlation significant at  $P \leq 0.05$  (probability of error max.5%); \*\*correlation significant at  $P \leq 0.01$  (probability of error max.1%); C2 – protein weakening; C3 – starch gelatinisation; C4 – stability of gel; C5 – starch retrogradation; C1C2 – fall of protein strength; C3C4 – diastatic activity; C5C4 – anti-stalling effect; DS – stability of the dough before weakening; DON – DON content; CP – crude protein; WG – wet gluten; ZS – Zeleny sedimentation; LV – loaf volume; H/D – rate of height and diameter of bread; FY – flour yield; AC – ash content

eters such as Zeleny sedimentation index or falling number as well as the main baking criteria – loaf volume and the shape features of the bread.

The complex rheological evaluation, low amount of the sample examined (about 45 g of flour), and fully automated analysis predestines the rheological system Mixolab for the prediction of the technological quality of flour in both food industry and breeding programmes.

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