

RESEARCH ARTICLE

(Open Access)**Fusarium infection degree and agrobiological properties of soft - wheat cultivars.**DORINA BELI^{1*}, ARTIONA LAZE¹, VJOLLCA GJINI², MIMOZA MUKAJ³, SKENDER VARAKU⁴^{1*}Agriculture University of Tirana, Faculty of Biotechnology and Food, Kamez, Tirane, Albania²The transfer Center of the Agriculture Technology, Lushnje, Albania³Albanian General Directorate of Customs, Tirana, Albania.

(mimoza.mukaj@yahoo.com)

⁴Agriculture University of Tirana, Faculty of Agriculture and Environment, Kamez, Tirane, Albania.*Corresponding author e-mail: dorinabeli9@gmail.com**Abstract**

Fusarium Head Blight of Wheat caused by *Fusarium spp* is one of the most widespread diseases in Albania which can cause significant yield losses if weather conditions are favorable (high humidity and temperatures above 24 degrees Celsius). This study was conducted to determine the influence of *Fusarium HeadBlight* index (Imc) on the quality of wheat grain .The following was analyzed : thousand kernel weight, crude protein content, wet gluten, ash content and k-SDS sedimentation in relation with disease index and internal infection on 13 winter wheat cultivars grown during the year 2015-2016 on experimental fields of Agriculture Technology Transfer Center (ATTC) Lushnja. Analysis of variance revealed significant variability's on total proteins, K-SDS and disease index (Imc) evaluating the susceptibility and the resistance of wheat cultivars against FHB . The statistical evaluation showed that there exists a medium negative correlation between thousand kernel weight and disease Index (Imc) ($r = -0.67$).

Key words: Fusarium Head blight, wheat , disease index, proteins, gluten.

Introduction

Wheat is often considered primarily as a source of energy (carbohydrate) and it is certainly important in this respect. However, it also contains significant amounts of other important nutrients including proteins, fiber, and minor components including lipids, vitamins, minerals, and phytochemicals which may contribute to a healthy diet. In particular, the unique properties of the gluten protein fraction allows the processing of wheat to produce bread and other baked goods and a range of functional ingredients[8] Winter wheat is one of the most important cereal crops in Albania. Kernel diseases of wheat are causing important losses of the yield. In Albania the total area planted with grain is 100.000 hectares almost half of the area planted before 1990. The total wheat production for 2014 was 280.000 tone (Albanian Statistical Institute INSTAT 2014).

Fusarium head blight (FHB) is a multifaceted disease caused by some species of *Fusarium spp*. A huge production of mycotoxins, mostly trichothecenes, often accompanied this disease. Amongst these toxic compounds, deoxynivalenol (DON) and its derivatives represent a major issue for human as well as for animal health and farming. Common and durum wheat are amongst the hosts of trichothecene-producing *Fusaria*. Infection from the fungus during the flowering stage of wheat can afflict starch qualities and it results in a higher degree of damaged starch [15] .This infection is responsible for a lower yield and a degradation of wheat quality [5] Weather conditions can be a limitation factor for wheat production due to the production of mycotoxine like trichothecenes which are not degraded during milling at high temperature treatment [17] . Mycotoxins are secondary metabolites produced by a wide variety of fungal species that cause nutritional

losses and represent a significant hazard to the food chain. The exposure risk to human is either directly through foods of plant origin (cereal grains) or indirectly through foods of animal origin (kidney, liver, milk and eggs) [16]. Maximum level of DON according to EU standards (EC2006) was set at 1250 ppb for unprocessed grain and 750 ppb for cereal flours. *Fusarium spp* destroys starch granules and storage proteins.[3] Damaged naturally in grain and it is an important factor during dough formation and this changes the absorption capacity, which increases from 30% to 100% in damaged starch. Also it has a high susceptibility against amylase activity. This could be a negative factor during the bread making process because during the infection *Fusarium spp* produces a large amount of amylase and it can lead to production of sticky and weak dough [14]. *F. graminearum* infection can cause significant changes in carbohydrate, lipid and protein composition. Cereals contaminated by *Fusarium spp* and their mycotoxins (DON), are not only a risk to human and animal health but also show reduced technological properties.[2] Additional changes also seriously affected the quality of storage proteins and dough properties [4]. Many authors emphasize the negative impact of different degrees of infection on bread quality but there are some studies which conclude that *Fusarium* infection does not alter the properties of dough formation [1]. The aim of this study is to correlate the effect of different infection grades with agro-biological parameters of wheat (ash, moisture, thousand kernel weight, protein, K-SDS sedimentation) which are important for the good quality of bread. The experiment was set on naturally contaminated wheat.

Material and methods

Materials

The effect of different infection grades of *Fusarium spp* was evaluated for 13 winter wheat cultivars samples planted during the years 2015-2016 on experimental fields of the State of Seed and

Saplings (ATTC) in Lushnje. The plant material was naturally contaminated by *Fusarium* infection during growth in the field. Field trials were carried out by methods of randomized blocks in four replications; the size of each plot was 12 square meters. For visual estimation for each plot, 10 heads were taken, at five places of, its diagonals, resulting in a total of 50 heads per plot.

2.2 Disease evaluation

Visual estimation of disease severity from natural infection *Fusarium spp* was used. Assessment of infection was based on standard area diagrams (SADs), the percent of the kernels surface occupied by the disease. Severity and diffusion of infection were obtained by resorting to the McKinney index [10]. The McKinney index (Imc) was obtained by using the following formula:

$$DI (Imc) = (f \times v) / N \times X \times 100$$

where: f = infection class frequencies; v = number of plants of each class N = total plants observed; X = highest value of the evaluation scale.

2.3 Chemical and technological analyses.

Moisture content was analyzed using the AOAC (1995) method 44 – 01. Crude protein was analyzed content (CP) according to Kjeldahl method (N x 5.7). The Wet gluten content (WG) in grain dry matter was determined using Glutomatic Perten, was analyzed according to AACC (2000) method 38-12.02. The sedimentation value (K-SDS) was determined according to Zeleny [18].

Results and discussion

The chemical – technological parameters of 13 cultivars of wheat grain are presented in Table 1. In the 13 analyzed cultivars the scale of infection ranges from high to moderate.

The moisture content within the analyzed cultivars ranged from 10.50% (Krajlica) to 12.30% (Bisancia). Thousand kernel of wheat (TKW) ranged from 36.23 g (LVS) to 48.98 g (Lucia). The highest reduction of TKW was genotype LVS (36, 23 g) and

Dajti (36.99 g). A higher TKW and higher test weight means a greater proportion of endosperm in the seed and thus a better yield of flour [13]. In recent studies the infection grade did not affect in particular the crude protein content [12]. According to Boyacioglu and Hettiarachy [3] the protein content increases significantly during a severe infection. In this study, it

was observed that there was a decrease in the value of protein content which ranged from 10.60% (Michelangelo) to 14.36% (Apache) which is also in accordance with the study by Gartner et al, who observed a decrease in protein content after severe *Fusarium* infection of grain [6]

Table 1. The chemical- agro-biological parameters of 13 cultivars

Cultivars	Moisture (%)	TKW (1000 Kernels weight) g	Imc (%)	K-SDS (ml)	Proteins (%)	WG (%)	Infection (%)	Ash %
Lucia	11.4	48.98	4.13	40.4	12.6	22	6.6	1.85
Mia	11.32	45.52	15.07	33	13.07	19.5	3.3	1.6
Katerina	11.85	42.5	11.77	30.2	14.96	20	33	2.18
Krajlica	10.5	37.24	16.8	29.7	13.9	10.6	9.9	2
Mateo	11.48	41.44	5.25	37.9	13.8	24.9	0	1.44
Europa	11.8	43.12	5.9	39.8	11.1	22	3.3	1.65
Apache	10.8	40.28	21.73	28.4	14.36	20.1	13.2	2.1
Simonid	11.19	41.86	33.6	29.3	12.82	15	6.6	2.2
Azul	11.6	42.76	4.07	40.2	10.6	24	3.3	1.54
Bisancia	12.3	43.07	28.57	34.7	12.7	23	3.3	1.98
Michelangelo	11.34	43.44	4.66	36.3	12.6	23.5	3.3	1.52
LVS	11.7	36.23	33	33.9	11.7	22	6.6	1.96
Dajti	11.44	36.99	32.27	29.5	14.07	16.1	13.2	2.16

K-SDS measures the swelling potential of the kernel proteins and in our cultivars it ranges from 29.50 ml (Dajti) to 42.40 ml (Lucia). According to Pisulewska.E et al (2011) [13] a general reduction of K-SDS after FHB infection was observed. Although the total amounts of proteins remains stable, FHB alters the quality of proteins [5][11]. The wet gluten test measures the amount of gluten protein in flour. According to, Laze et al 2015, the content of wet gluten in Albanian undamaged cultivars ranges from 23.43% to 33.20%, with an average value of 28.94%. [9]. Other studies found wet gluten content reduced in artificially *Fusarium* infected samples.[3],[13],[11]. In the analyzed samples, specifically Dajti (DI=32.27%), and Simonid (DI=33.6%), there was a decline in wet

gluten content as well as a high rate of infection. Also the handling properties of wet gluten seemed to be moderately affected and the wet gluten was very sticky and difficult to handle.

Thousand kernels weight was negatively correlated with internal infection ($r = -0.61$) and Imc ($r = -0.67$), while Imc showed a negative correlation with K-SDS ($r = -0.694$) (Table 2). This is in accordance with many studies that suggest that FHB has a negative impact on yield and protein quality [13]. The most susceptible cultivar Dajti (DI =32.27) had the lowest 1000 kernel weight and the lowest value for K-SDS (29.5ml) [13]. An increase of ash content of flour was recorded in the most infected cultivars. Ash is mainly composed of minerals of the

seed coat (bran and aleurone), the proportion of the ash in flour is an indicator of its purity. An increase in ash content means an alteration on the kernel seed coat volume ratio. It is possible to associate this estimation with the presence of shriveled *Fusarium* damaged kernels [7].

According to the following study, results indicated that the cultivars Lucia, Mateo and with

small mean values of the McKinney's index DI (Imc), have shown results of resistance (R) against *Fusarium spp.*. Dajti and LVS (control) with mean values of the McKinney's index DI (Imc), have shown a moderate resistance (MR) level against *Fusarium spp.* (Table 1).

Table 2. Correlation coefficient between chemical-agro-biological parameters

	Moisture (%)	TKW (thousand kernel weight)	intern inf. (%)	Imc (%)	K-SDS (ml)	Protein (%)	WG (%)
Moisture (%)	1						
TKW (g)	0.21	1					
Intern inf. (%)	-0.3	-0.1	1				
Imc (%)	-0.14	-0.67	0.1	1			
K-SDS (ml)	0.43	-0.07	-0.55	-0.69.4	1		
prot (%)	-0.13	-0.21	0.28	0.51	0.1	1	
WG (%)	0.62	0.3	-0.35	-0.03	0.72	-0.19	1

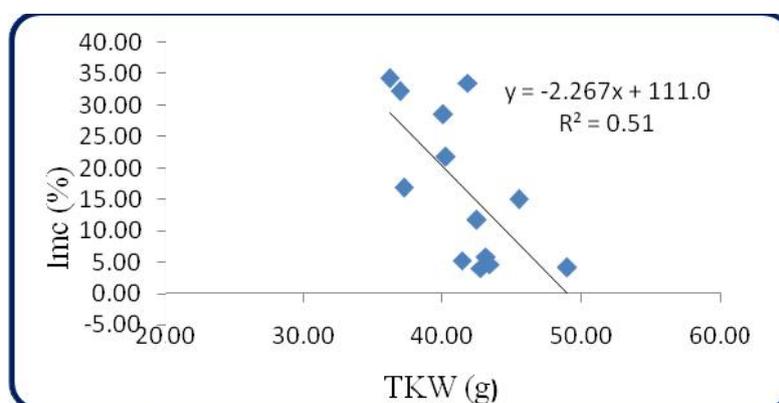


Figure 1. Linear regression of TKW(g) with Imc (%)

Using a linear regression analysis, a lower negative correlation between Imc (%) and thousand kernel weight was determined (Figure 1). These correlations can be described by the equation as follows: $Imc (\%) = 111.0 - 2.267 * \text{Thousand kernel weight}$.

The data was analyzed by one way ANOVA (single factor) using excel 2010 for significant F-statistics, if the overall F-test was significant ($F < 0.05$) a Fachers T-test was performed to discern differences between the varieties.

Table 2. ANOVA analyses for the disease index

<i>Source of Variation</i>	<i>SS</i>	<i>df</i>	<i>MS</i>	<i>F</i>	<i>P-value</i>	<i>F crit</i>
Between Groups	5225.48	12	4.35.46	945.55	3.95E-31	2.15
Within Groups	11.97	26	0.46			
Total	5237	38				

Table 3. ANOVA analyses for the protein content

<i>Source of Variation</i>	<i>SS</i>	<i>df</i>	<i>MS</i>	<i>F</i>	<i>P-value</i>	<i>F crit</i>
Between Groups	62.55	12	5.21	42.82	1.18E-18	2.01
Within Groups	4.75	39	0.12			
Total	67.29	51				

Table 4. ANOVA analyses for wet gluten

Source of Variation	SS	df	MS	F	P-value	F crit
Between Groups	587.67	11	53.42	312.73	4.05987E-23	2.22
Within Groups	4.10	24	0.17			
Total	591.77	35				

Table 5. ANOVA analyses for K-SDS

<i>Source of Variation</i>	<i>SS</i>	<i>df</i>	<i>MS</i>	<i>F</i>	<i>P-value</i>	<i>F crit</i>
Between Groups	646.95	12	53.91	50.63	6.61E-15	2.15
Within Groups	27.69	26	1.06			
Total	674.64	38				

Analyses of variance revealed significant differences between the cultivars for all of the biochemical and chemical traits (protein content, sedimentation value, wet gluten content and Imc) (Table 2, 3, 4, 5). The difference between cultivars may be due to variation in their genetic variability.

Conclusions:

This study demonstrates the effect of FHB in certain biochemical components of wheat. The total amount of crude proteins has a light increase during the infection of *Fusarium spp* as well as a decrease in the quality of the storage proteins, according to K-SDS value which decreases significantly after the FHB infection in most susceptible cultivars. The

cultivars Mia, Lucia, Mateo and Michelangelo showed the best performance in confronting the disease index but medium values for K-SDS and total protein content. Analysis of variance revealed the significant variability in the experimental materials used to evaluate the susceptibility and the resistance of common wheat cultivars against FHB. The statistical evaluation showed that there exists a medium negative correlation between thousand kernel weight and disease Index (Imc) ($r = -0.67$).

References :

1. Antes S, Birzel B, Prange A, Krämer J, Meier A, Dehne H.W, Köhler P., **Rheological and bread-making properties of wheat samples infected**

- with *Fusarium* spp. 2001. *Mycotoxin Res.*, 17:76-80.
2. Birzele B., Prange A: **Fusarium-inoculated wheat. Deoxynivalenol contents and baking quality in relation to infection time.** *Food Control.* 2005. 739-745.
 3. Boyacio lu D, Hettiarachchy N.S. **Changes in some biochemical components of wheat grain that was infected with *Fusarium graminearum*.** *J. Cereal Sci.*, 1995. 57–62.
 4. Dexter J.E, Clear R.M, Preston K R. **Fusarium head blight effect on the milling and baking of some Canadian wheats.** *Cereal Chem.* 1996. 695–701.
 5. Eggert K, Rawel, H.M., Pawelzik, E: **In vitro degradation of wheat gluten fractions by *Fusarium*.** *Eur Food Res Technol.* 2011. 233:697–705.
 6. Gärtner B.H, Munich M, Kleijer G, Mascher F. **Characterisation of kernel resistance against *Fusarium* infection in spring wheat by baking quality and mycotoxin assessments.** *Eur. J. Plant Pathol*, 2008. 120: 61–68.
 7. Horvat D, Spanic D. : **The Influence of *Fusarium* Infection on Wheat (*Triticum aestivum* L.) Proteins Distribution and Baking Quality.** *Cereal Res. Commun.* 2015. 43: 61–71.
 8. Kumar R, Yadava B, Gollen S , Verma RK, Yadav S. **Nutritional Contents and Medicinal Properties of Wheat: A Review.** *Life Sci. Med. Res.*2011: LSMR-22.
 9. Laze A, Arapi V, Malo V, Kristil J, Grobelnik S, Pezo L. **The qualities of Albanian Soft wheat Genotypes – the Mathematical Approach.** *Int. J. Sci Qual Anal.* 2015. 1: 11- 17.
 10. McKinney H. H., **Influence of soil temperature and moisture on infection of wheat seedling by *Helminthosporium sativum*.** *J.Agric.Res.* 1923. 26:195 – 217.
 11. Nightingale M.J , Marchylo B.A , Clear R.M , Dexter J.E., Preston K.R: **Fusarium head blight: Effect of fungal proteases on wheat storage proteins.** *Cereal Chem*, 1999. 76: 150–158.
 12. Papoušková L., Capouchová I., Kostelanská M., Ške íková A., Prokinová E., Hajšlová J., Salava J., and Fam ra O.; **Effect of different intensities of *Fusarium* infestation on the grain yeld ,deoxynuivalenol content and baking quality of winter wheat.,** *Rom Agric Res.* 2012, 305-306.
 13. Pisulewska E, Torbica A, Mastilovic J. **Influence of degree of wheat infestation with *Fusarium* on his technological quality and safety.** *Proc. Nat. Sci, Matica Srpska Novi Sad*, 2011. 120, 61—70.
 14. Hamer R.J and Hoseneý R.C. **Interactions: The Keys to Cereal Quality. AACCI 1998.**
 15. Scala V.,Aurelia G., Cesarano G. **Climate, Soil Management, and Cultivar Affect *Fusarium* Head Blight Incidence and Deoxynivalenol Accumulation in Durum Wheat of Southern Italy.** *Front Microbiol.* 2016, 7: 1014.
 16. Yazor S, Omurtag.G; **Fumonisin, Trichothecenes and Zearalenone in Cereals.** *Int J Mol Sci.* 2008. 2062–2090.
 17. Zain.E.M. **Impact of mycotoxins on humans and animals.** *J. Saudi Chem. Soc.* 2011. 129–144.
 18. Zelený L. A. **Simple sedimentation test for estimating the bread – baking and gluten qualities of wheat flour.** *Cereal Chem.* 1947. 24, 465–475.