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MULTI-ENVIRONMENT EVALUATION OF DISEASE OCCURRENCE, AGGRESSIVENESS AND WHEAT RESISTANCE IN WHEAT/*FUSARIUM* PATHOSYSTEM

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ABSTRACT

Improvement of assessment methods and selection of new robust sources of resistance, are wheat pre-breeding goals of outmost importance at NARDI Fundulea. Natural occurrence of FHB/DON contamination, host resistance and pathogen aggressiveness in current *Triticum aestivum*/*Fusarium* pathosystems under artificial inoculation were investigated. Analysis of 715 wheat samples collected from natural conditions revealed up to 10% risks of fungus contamination in three out of 15 environments, locations Livada (N/W Romania) and Piteşti-Albota (S/W Romania), being relatively the most vulnerable to FHB occurrence. Level of DON accumulation in grain was generally under 1.25 mg/kg, the limit reinforced for grain by UE regulations. A broad range of variation for aggressiveness in seedling and adult stages among 30 and 10 respectively, *Fusarium* accessions collected from five locations was found. Based on varietal response to FHB inoculation performed with both, point and spray inoculation methods, phenotypic differences between resistance types of 20 wheat Romanian cultivars were noticed that suggest association of resistance Type 1 and 2 (Dumbrava, Delabrad, Dor) or prevalence of Type 1 (Izvor, Crina, Magistral).

Key words: wheat resistance, *Fusarium* aggressiveness, pathosystem, DON contamination**.**

INTRODUCTION

The semi-biotrophic pathogen *Fusarium*, primarily *F. graminearum* Schwabe [(teleomorph *Gibberella zeae* Schw. (Petch)] is the cause of head blight (FHB, scab), a serious threat to the worldwide grain industry in favorable conditions for development of epidemics. Beyond yield reduction, FHB is a detrimental factor for food safety and consumers health because of potential grain contamination with its associated mycotoxins including deoxynivalenol (DON). There are contradictory reports in the literature concerning the close correlations between FHB and DON content (Semagn et al., 2007), 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). Cleaning and removing kernels with low specific gravity or through optical seed-sorting technology can reduce DON contamination, but about 60% of it remains after milling, and not appreciable loss of toxins was detected after baking (Young et al., 1984; Seitz et al., 1986; Tkachuk et al., 1991; Tanaka et al., 1999, cited by Nishio et al., 2010).

While crop management practices and chemical applications may reduce the damages (Zhang et al., 2010), development and deployment of resistant cultivars, is unanimously considered as a key component in order to mitigate economically and environmentally friendly the various risks of FHB (Bai and Shaner, 2004; Buerstmayr et al., 2009), in both conventional and organic agronomic systems. Release of resistant cultivars has been hampered by several factors: 1) quantitative nature of FHB resistance, 2) complexity of both host resistance and pathogen aggressiveness and, 3) high genotype x environment interactions. However, in the last years a better knowledge on host/pathogen

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interactions with considerable impact on improvements of genetic resistance was achieved. Significant genetic resources for FHB resistance and their corresponding quantitative trait loci (QTL) for the known resistance types [(Type 1 = resistance to initial infection; Type II = resistance to spread of the fungus to adjacent spikelets *sensu* (Schroeder and Christensen, 1963) and Type III = resistance to mycotoxin contamination (Miller et Arnison, 1986)] have been reported from various gene pools originated from Asia, Europe, South and North America. QTL's were genetically mapped on almost all of the 21 wheat chromosomes in germplasm from many Asian, North American, South American, and European countries, *fhb1* located on *3BS* being the best validated (Van Sanford et al., 2001; Häberle et al., 2007; Buerstmayr et al., 2009; Liu et al., 2009) and used extensively in FHB breeding programs worldwide (Badea et al., 2008). Additionally in the recent years, a better explanation of factors involved in wheat grain contamination with mycotoxins has been registered (Lemmens et al., 2005; Ma et al., 2006). Studies of trichotecene genes *(Tri)* expression of *Fusarium graminearum* aimed to identify genetic variation of wheat resistance to the fungal toxic compounds became a new research approach. PCR markers for *Tri 7* and *Tri 13* genes in order to predict occurrence of new chemotypes of DON-producing strains of *F. graminearum* were developed (Chandler et al., 2003). Evolutionary rate of *Fusarium* populations across world, according to their genetic variation is considered rather low as compared with air transmitted pathogens of wheat (i.e.: rusts). However an adaptive evolutionary shift in FHB pathogen populations toward the rapid spread of more aggressive chemotypes (i.e. 3-acetyl DON, 3-ADON) as compared with 15-ADON, in terms of DON production has been reported in *F. graminearum* populations from North America (Starkey et al., 2008; Ward et al., 2008). Change of prevalence among the two analogs may dramatically influence current FHB management strategies.

As a consequence continuous selection of more aggressive *Fusarium* isolates and use of combined breeding methods for disease evaluation are prerequisite conditions in order to minimize the alteration of expressions of resistance, generally determined by genetic backgrounds of both host and pathogen, and by the strong influence of environment.

Development of FHB resistance in wheat germplasm and improvement of assessment methods, under field artificial inoculation, are pre-breeding goals of outmost importance at NARDI Fundulea. The aims of this study were to evaluate in various environments and development stages: 1) natural occurrence of FHB and DON contamination and 2) host resistance and pathogen aggressiveness in current *Triticum aestivum*/*Fusarium* pathosystems under artificial inoculation.

MATERIAL AND METHODS

FHB and DON occurrence

Investigations under natural conditions in six (2007 and 2008) and three locations (2009) across Romania, were performed. Level of *FHB* contamination in 195 to 264 random grain samples/year, and 25-79 entries/location/year, was analyzed. About 100 seeds/entry were planted on agar medium (20 g/l) in three replications. After 48 days at +25°C in dark the number of seeds showing grown *Fusarium* colonies was counted and the level of contamination was expressed as percentage (%). Estimation of DON content in wheat samples randomly harvested from 3-6 locations/year, was performed at the Research & Development Institute for Plant Protection, Bucharest, by ELISA on Ridascreen®FAST DON (ppm = mg/kg) kits (R-Biopharm GmbH, Darmstadt, Germany), according to the manufacturer's instructions.

Fusarium isolates

30 *Fusarium* accessions obtained at NARDI Fundulea from the naturally infected grains of bread wheat, durum wheat and triticale, sampled in 2008 from five locations (Piteşti-Albota, Livada, Braşov, Şimnic and Tg. Mureş) were analyzed in seedling stage, while for field experiments by point inoculation only ten selected *Fusarium* isolates of them and other five reference isolates were

used (Table 1). The high aggressive reference isolate F. c. 46, in culture over 28 years is intensively used for experimental assessment of resistance (Miedaner et al., 1996). *Fusarium graminearum* isolates F. g. 96, F. g. 8713, F. g. 735, and F. g. 771 were previously collected by the authors from Romanian wheat samples.

The prevalence of *F. graminearum* morphologically identified in this collection was confirmed by molecular tools (Petruța Cornea, *personal communication*). Inoculum necessary for artificial inoculations, consisting of homogenized suspensions of conidia in distilled water (about 500000/ml), was produced from *Fusarium* isolates cultured on the both, Czapek Dox medium and sterilized wheat grains, for the first 14 days at +25°C in dark and exposed to black light (Philips TLO, 40 W/80) for the following seven days. The same inoculum was sprayed twice, at beginning and mid anthesis at a rate of 250 ml/plot.

Table 1. Fusarium isolates analyzed for pathogen aggressiveness and host resistance under artificial inoculation in seedling and adult stage

Fusarium code	Origin			Method of inoculation (stage)		
				Seedling 2008	Adult (2008 and 2009)	
	Year	Location	Host		Point	Spray
F. c. 46 (IPO39-01)*	**1966**	**Holland**	**wheat**	**-**	**X**	**mixture**
F. g. 96	**1995**	**Braşov**	**wheat**	**-**	**X**	**mixture**
F. g. 8713	**1987**	**Fundulea**	**wheat**	**-**	**X**	**mixture**
F. g. 735	**2007**	**Livada**	**wheat**	**-**	**X**	**mixture**
F. g. 771	**2007**	**Piteşti-Albota**	**wheat**	**-**	**X**	**mixture**
F. g. 983	2008	Brasov	durum	X	-	-
F. g. 991	2008	Brasov	durum	X	-	--
F. c. 1056	2008	Simnic	wheat	X	X	-
F. g. 1145	2008	Piteşti-Albota	wheat	X	X	-
F. g. 1137	2008	Piteşti-Albota	wheat	X	-	-
F. g. 1143	2008	Piteşti-Albota	wheat	X	-	-
F. g. 1156	2008	Piteşti-Albota	wheat	X	-	--
F. g. 1182	2008	Piteşti-Albota	wheat	X	-	-
F. g. 1169	2008	Piteşti-Albota	triticale	X	-	-
F. g. 1204	2008	Tg.Mureş	wheat	X	X	-
F. g. 1211	2008	Tg.Mureş	wheat	X	X	-
F. g. 1224	2008	Livada	wheat	X	X	-
F. g. 1228	2008	Livada	wheat	X	X	-
F. g. 1237	2008	Livada	wheat	X	X	-
F. g. 1239	2008	Livada	wheat	X	X	-
F. g. 1265	2008	Livada	wheat	X	X	-
F. g. 1272	2008	Livada	wheat	X	X	-
F. g. 1216	2008	Livada	wheat	X	-	-
F. g. 1220	2008	Livada	wheat	X	-	
F. g. 1226	2008	Livada	wheat	X	-	-
F. g. 1274	2008	Livada	wheat	X	-	-
F. g. 1278	2008	Livada	wheat	X	-	-
F. g. 1222	2008	Livada	wheat	X	-	-
F. g. 1238	2008	Livada	wheat	X	-	-
F. g. 1266	2008	Livada	wheat	X	-	--
F. g. 1313	2008	Livada	triticale	X	-	-
F. g. 1316	2008	Livada	triticale	X	-	--
F. g. 1317	2008	Livada	triticale	X	-	-
F. g. 1318	2008	Livada	triticale	X	-	-
F. g. 1343	2008	Livada	triticale	X	-	-

Reference *Fusarium* isolates (bolded) *) By the courtesy of Thomas Miedaner (University of Hohenheim, Germany)

Pathogenicity test

Aggressiveness, in seedling and adult stage, at NARDI Fundulea was estimated. In seedling stage, aggressiveness of *Fusarium* isolates *vs*. three wheat entries was evaluated, according to the protocol established by Ittu (1986). Pathogenic potential of *Fusarium* isolates was expressed as reduction of coleoptile length in eight days old seedlings. In adult stage aggressiveness was analyzed in 190 host x pathogen combinations: five reference *Fusarium* isolates x 20 wheat cultivars and ten *Fusarium* isolates x nine wheat cultivars, respectively.

Resistance trials

Resistance of 20 modern Romanian varieties (Mustăţea et al., 2009), released between 1991-2009, by NARDI Fundulea and the wheat breeding centers from Piteşti, Lovrin, Turda, Suceava and Podu Iloaiei (Table 2), was assessed by point and spray artificial inoculations with a mixture of ten and respectively, five *Fusarium* isolates in the FHB experimental field at Fundulea. Level of host resistance/pathogen aggressiveness in adult stage was phenotipically analyzed in terms of severity (damaged florets at 20 days post inoculation, %), area under disease progress curve (AUDPC) and *Fusarium* damaged kernels, FDK %.

Table 2. Romanian wheat varieties analyzed for resistance to FHB under artificial inoculation (Fundulea, 2008-2009)

Variety	Released		Cultivated area in 2009, in Romania (%)
	Breeding center	Year	
Dropia	NARDI Fundulea	1993	19.85
Alex	NARDI Fundulea	1994	11.29
Boema	NARDI Fundulea	2000	19.84
Crina	NARDI Fundulea	2001	2.05
Dor	NARDI Fundulea	2002	0.76
Delabrad	NARDI Fundulea	2002	2.48
Faur	NARDI Fundulea	2004	0.11
Glosa	NARDI Fundulea	2005	7.29
Gruia	NARDI Fundulea	2005	0.52
Izvor	NARDI Fundulea	2009	0.10
Albota	ARDS Piteşti-Albota	1986	0.10
Trivale	ARDS Piteşti-Albota	1991	0.16
Briana	ARDS Şimnic	2004	0.30
Ciprian	ARDS Lovrin	2003	1.37
Arieşan	ARDS Turda	1985	2.12
Apullum	ARDS Turda	1992	0.33
Dumbrava	ARDS Turda	2003	0.62
Magistral	ARDS Suceava	1998	0.24
Gaşparom	ARDS Suceava	1998	0.62
Beti	ARDS Podu-Iloaiei	2004	0.01

Statistical analysis

Analysis of variance (ANOVA) was used to estimate the contributions attributable to genotypes of the pathogen and host, environment and method of inoculation. Correlation analysis was calculated to estimate relationships between resistance traits over years and inoculation methods.

RESULTS AND DISCUSSION

Climatic conditions

Artificial spray inoculations were performed during anthesis, between days (after January 1) 127 and 147 (2008) and, from 135 to 141 (2009), respectively. Average temperatures (°C) and total rainfall (mm), the main climatic factors of concern for development of FHB in natural conditions were recorded 20 days post inoculation (d.p.i.), the period considered as critical for disease expression. In 2009 inoculations started 2-6 days earlier as compared to 2008, higher differences among environments being registered particularly regarding to rainfall. So, during the inoculation interval, from day 137 to day 142 (in 2009), less moisture (3.1-6.4 mm) was available, while during the inoculation period from day 144 to 147, rainfall was 8.1-14.3 mm higher than in 2008 (Table 3).

Occurrence of FHB and DON contamination

Analyses of wheat grains sampled from 15 environments showed on average a variation of grains (%) infected with *Fusarium* ranging from 1.9 % (2007) to 6.2 % (2009). Based on these results, a relatively higher risk of contamination could be assumed at Livada (N/W of Romania), where values higher than

9.0 % in 2008 and 2009 were observed, while at Piteşti-Albota (S/W of Romania) in only one of three years, a FHB attack of 7.7% was determined. At Fundulea (South Romania), no evidence of significant natural occurrence of FHB was noticed, but for a better characterization of disease occurrence in other Romanian centers favorable for wheat crop, more consistent investigations are necessary (Table 4).

Table 3. Average temperature and total rainfall during 20 postinoculation days

Varieties	Inoculation day (after January 1st)			Average temperature (°C)			Rainfall (mm)		
	2008	2009	*Diff.*	2008	2009	*Diff.*	2008	2009	*Diff.*
1. Briana, Glosa	137	135	*-2*	18.9	19.5	*0.6*	27.6	24.5	*-3.1*
2. Gruia, Dropia	140	138	*-2*	18.6	19.4	*0.8*	27.7	21.3	*-6.4*
3. Boema, Izvor, Ciprian, Crina, Trivale, Arieşan	142	138	*-4*	18.6	19.6	*1.0*	26.2	21.3	*-4.9*
4. Faur, Alex, Dor, Beti, Delabrad, Albota	144	139	*-5*	19.2	19.6	*0.4*	7.0	21.3	*14.3*
5. Apullum, Gaşparom	145	138	*-7*	19.4	19.4	*0*	13.2	21.3	*8.1*
6. Magistrala, Dumbrava	147	141	*-6*	19.4	20	*0.6*	13.2	24.6	*11.4*

Table 4. Natural occurrence of *Fusarium* head blight in wheat samples collected in 15 environments (% of infected grains, mean values)

Location	2007		2008		2009		Average	
	samples	% attack	samples	% attack	samples	% attack	samples	% attack
Fundulea	25	0	42	0	45	1.1	37	0.4
Piteşti-Albota	50	1.2	49	1.1	110	7.7	70	3.3
Livada	45	3.3	57	9.3	40	9.8	47	7.5
Tg. Mureş	40	1.0	31	1.4	-	-	36	1.2
Braşov	79	3.0	42	1.1	-	-	60	2.1
Şimnic	-	-	35	0.2	-	-	35	0.2
Secuieni	25	3.0	-	-	-	-	25	3.0
Total samples	264	-	256	-	195	-	715	-
Average		1.9		2.2		6.2		3.4

Estimation of DON content in wheat grain samples that were analyzed suggested a rather reduced risk of contamination with this mycotoxin, according to European limits of DON level in wheat grains [Commission Regulation (EC) no 1126/2007].

Values exceeding UE regulated content of 1.25 ppm = 1.25 mg/kg were found only as an exception in a sample from Livada (2009), in which 1.38 ppm of DON was quantified. Values higher than 1.0 ppm were found mainly in Piteşti-Albota (2007 and 2008) (Table 5).

The higher percentage of FDK found in both locations confirms previous findings that this parameter could be generally considered as a good indicator of DON contamination (Miedaner et al., 2004; Ittu, 2006).

Table 5. Contamination with FHB (%) and DON (ppm) content in 15 environments (maximum values)

Location	2007		2008		2009		Average	
	FDK, %	DON (ppm)	FDK, %	DON (ppm)	FDK, %	DON(ppm)	FDK, %	DON(ppm)
Fundulea	0	0.28	0	0.54	10.0	0.12	3.3	0.3
Piteşti-Albota	13.3	1.10	7.5	1.02	27.5	0.15	16.1	0.8
Livada	13	0.24	30	1.38	47.5	0.14	30.2	0.6
Tg. Mureş	6	0.10	17.5	0.37	-	-	11.8	0.2
Braşov	16.6	0.08	20	0.01	-	-	18.3	0.05
Şimnic	-	-	2.5	1.13	-	-	2.5	1.13
Secuieni	13.3	0.37	-	-	-	-	13.3	0.37
Average	12.4	*0.38*	15.5	*0.78*	25.0	*0.17*	15.4	*0.52*

Pathogen aggressiveness

A broad range of aggressiveness variation among *Fusarium* accessions in seedling stage was found. Reduction of coleoptile length, varied on average from 2.1 to 30.9% as compared with control. Relatively highly aggressive *Fusarium* isolates that produced a reduction of coleoptiles length exceeding 25.0% as compared with control, were obtained from naturally infected samples originated from Piteşti-Albota (F. g 1156, F. g. 1182), Livada (F. g. 1272, F. g. 1226, F. g. 1228) and Şimnic (F. c. 1056) (Figure 1).

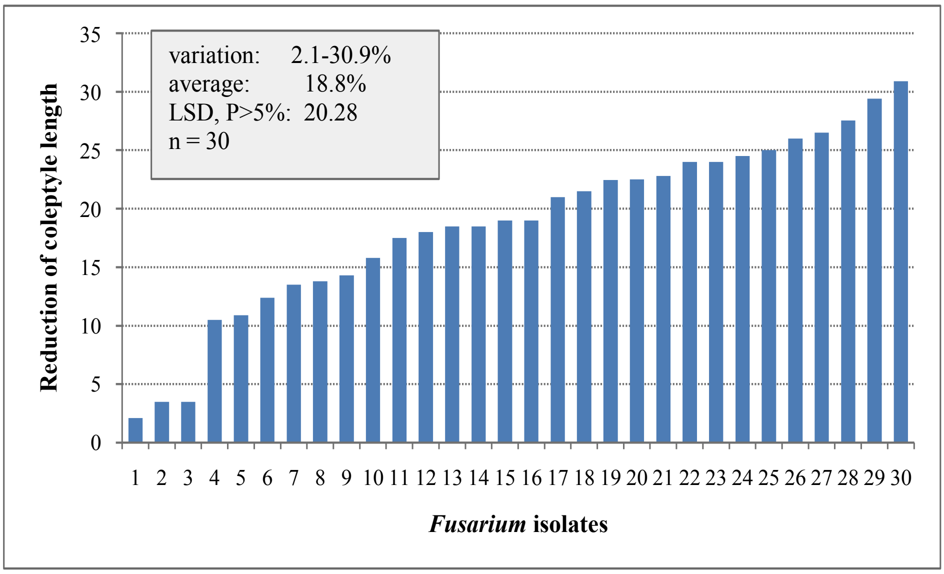

Figure 1. Estimation of *Fusarium* aggressiveness in seedling stage (Fundulea, 30 isolates, mean values)

Fusarium isolates: 1) F.g. 1266; 2) F.g. 1216; 3) F.g. 1143; 4) F.g. 1313; 5) F.g. 983; 6) F.g. 1220; 7) F.g. 1169; 8) F.g. 1318; 9) F.g. 991; 10) F.g. 1222; 11) F.g. 1137; 12) F.g.1237; 13) F.g. 1278; 14) F.g. 1238; 15) F.g. 1317; 16) F.g. 1316; 17) F.g. 1343; 18) F.g. 1224; 19) F.g. 1211; 20) F.g. 1274; 21) F.g. 1145; 22) F.g. 1239; 23) F.g. 1204; 24) F.g. 1265; 25) F.g 1156; 26) F.g. 1182; 27) F.g. 1272; 28) F.g. 1226; 29) F.g. 1228; 30) F.c. 1056.

Table 6. Components of aggressiveness under field point inoculation
(Fundulea 2009, ten isolates *vs*. nine wheat varieties, mean values)

Entry	Severity, %			AUDPC			FDK, %		
	Minimum	Maximum	Average	Minimum	Maximum	Average	Minimum	Maximum	Average
F. c. 1056	28.3	92.4	61.8	217.5	707	527.1	10.4	82.3	43.7
F. g. 1265	11.4	75	42.8	89.5	536	312	1.9	59.1	30.5
F. g. 1224	31.6	100	64.8	231	800	489	0.8	39.4	29
F. g. 1272	15.1	65.7	44.3	145.5	462.5	321.3	0.7	46.5	27.5
F. g. 1237	2.6	68.5	48.9	16	464.5	329.6	0	41.4	26.6
F. g. 1145	20.8	62.7	46.9	193	458.5	342	6.3	49	26
F. g. 1211	22.6	79.7	54.1	159	753	439.9	3.5	51	26.0
F. g. 1204	16.5	57.3	39.7	113.5	437	298.4	3.2	54.6	25.7
F. g. 1228	15	82.3	40.7	147	593.5	326.6	5	38.5	22
F. g. 1239	8.3	23.9	14.4	72	145.5	104.9	0	18.3	8.1
Average	*17.2*	*70.8*	*45.8*	*138.4*	*535.8*	*349.1*	*3.2*	*48.0*	*26.5*
Minimum	*2.6*	*23.9*	*14.4*	*16*	*145.5*	*104.9*	*0*	*18.3*	*8.1*
Maximum	*31.6*	*100*	*64.8*	*231*	*800*	*527.1*	*10.4*	*82.3*	*43.7*
LSD for P<5%			11.91			79.86			9.99

In adult stage a large variation of aggressiveness in terms of severity (%), AUDPC and FDK %, among the ten analyzed *Fusarium* isolates *vs.* nine wheat cultivars was found. The corresponding limits of variation for these parameters were on average 14.4-64.8% (severity), 104.9-527.1 (AUDPC), 8.1-43.7% (FDK). Similarly to the experiment carried on in seedling stage, *Fusarium culmorum 1056* isolated from Şimnic (entry number 30) expressed in adult stage the highest level of relative aggressivity in respect of AUDPC (527.1) and FDK, % (43.7) (Table 6). In this case, phenotypic records were in agreement with genotyping data (Petruța Cornea, *personal communication*).

Host resistance

Data analysis showed that coefficients of correlation between methods of inoculation/year combinations varied, as expected, according to climatic differences registered among years, being low for spray inoculation regardless of FHB resistance trait, severity, % (0.26), AUDPC (0.034), and FDK, % (0.33), respectively (Tables 7-9). Significant high correlations (P<0.1) were found for the relationship between results of point inoculation over years for % severity (*r* = 0.55), AUDPC (*r* = 0.63) and % FDK (*r* = 0.60) and on average over years between the two inoculation methods. With regard to FDK, very highly significant correlation (P<0.01) between results of point and spray inoculation methods was found (*r* = 0.76) (Table 9, Figure 2) that suggests a higher stability of this trait over environments.

Table 7. Coefficients of correlation (*r* values) for severity (%) estimated over methods of inoculation x environments combinations

Method of inoculation/year	Spray 2009	Point 2009	Spray (average)
Spray 2008	0.26		
Point 2008		0.55**	
Point (average)			0.59**

***Significantly different from zero at P<0.001
** Significantly different from zero at P<0.01
* Significantly different from zero at P<0.05

Table 8. Coefficients of correlation (*r* values) for AUDPC (%) estimated over methods of inoculation x environments combinations

Method of inoculation/year	Spray 2009	Point 2009	Spray (average)
Spray 2008	0.034		
Point 2008		0.63**	
Point (average)			0.47*

***Significantly different from zero at P<0.001
** Significantly different from zero at P<0.01
* Significantly different from zero at P<0.05

Table 9. Coefficients of correlation (*r* values) for FDK (%) estimated over methods of inoculation x environments combinations

Method of inoculation/year	Spray 2009	Point 2009	Spray (average)
Spray 2008	0.33		
Point 2008		0.60**	
Point (average)			0.76***

***Significantly different from zero at P<0.001
** Significantly different from zero at P<0.01
* Significantly different from zero at P<0.05

Variation of severity (%), AUDPC, and FDK (%), was lower for the late cultivars as compared to the earlier ones. The lowest values of FDK (<10%) were found in cultivars Dumbrava (5.9), Delabrad (7.9) and Dor (9.0), while other nine cultivars expressed relatively moderate resistance according to this parameter (<20%) (Table 10).

Regression of FDK (%) varietal response to spray inoculation on the response to point inoculation revealed a higher level of resistance mainly in cultivars Dumbrava, Delabrad and Dor, regardless of the applied method of inoculation.

The cultivar Gruia, was more susceptible to spray inoculation, as compared to point inoculation. Based on data obtained for varieties Izvor, Crina and Magistral, resistance Type 1 seems to be prevalent in these varieties.

Albota, Trivale and Arieşan were the most susceptible entries to FHB in this study (Figure 2). These findings could suggest differences among Romanian varieties in respect to their characteristic FHB resistance types, known as being phenotipically distinguishable by spray inoculation (type I) and point inoculation (Type II), respectively (Bai and Shaner, 1996).

Understanding the pattern of host response to pathogens is a core requirement for enabling host-pathogen system specific disease management. Complexity of resistance-aggressiveness and strong interference with environment emphasized this demand in breeding of wheat resistance vs. *Fusarium* fungus.

According to Bai and Shaner (2004), adding type I resistance into lines with type II resistance should increase their field resistance against FHB.

Table 10. Components of resistance to FHB under point and spray field inoculation across two environments and 6 inoculation intervals.
(Fundulea 2008-2009, 20 varieties *vs.* five reference *Fusarium* isolates, mean values)

Inoculation Interval	Variety	FHB resistance traits								
		Severity			AUDPC			FDK		
		Point	Spray	*Average*	Point	Spray	*Average*	Point	Spray	*Average*
1	Briana	41.93	11.16	*26.5*	334.4	163.1	*248.7*	16.5	26.8	*21.6*
	Glosa	39.46	6.93	*23.2*	346.9	112.1	*229.5*	14.7	29.9	*22.3*
2	Gruia	31.54	12.82	*22.2*	257.7	229.8	*243.7*	9.2	26.7	*17.9*
	Dropia	35.37	10.21	*22.8*	282.9	166.4	*224.6*	13.7	34.6	*24.1*
3	Boema	39.37	6.685	*23.0*	323.2	128.9	*226.0*	14.1	32.2	*23.1*
	Izvor	32.33	6.12	*19.2*	289.9	207.6	*248.7*	13.8	12.2	*13.0*
	Ciprian	37.73	9.71	*23.7*	283.2	265.4	*274.3*	14.5	19.3	*16.9*
	Crina	32.95	5.09	*19.0*	270.7	167.1	*218.9*	14.6	14.9	*14.8*
	Trivale	39.52	9.08	*24.3*	354.7	284.5	*319.6*	47.4	34.9	*41.1*
	Arieşan	40.4	14.04	*27.2*	376.8	275.9	*326.4*	23.8	52.9	*38.3*
4	Faur	32.62	4.825	*18.7*	265.1	123.8	*194.4*	10.7	15.6	*13.1*
	Alex	35.28	6.95	*21.1*	310.8	217.5	*264.1*	11.0	13.7	*12.4*
	Dor	24.12	4.63	*14.4*	213.5	130.2	*171.9*	9.8	8.1	*9.0*
	Beti	27.66	7.89	*17.8*	226.5	267.6	*247.0*	15.0	27.9	*21.4*
	Delabrad	21.44	5.78	*13.6*	197.2	156.2	*176.7*	5.6	10.2	*7.9*
	Albota	37.49	9.39	*23.4*	311.2	228.0	*269.6*	20.3	38.3	*29.3*
5	Apullum	41.64	13.2	*27.4*	372.8	387.2	*380.0*	16.5	22.5	*19.5*
	Gaşparom	35.8	7.89	*21.8*	314.6	225.2	*269.9*	11.5	18.3	*14.9*
6	Magistral	31.65	4.775	*18.2*	276.0	134.5	*205.2*	15.2	9.6	*12.4*
	Dumbrava	18.17	5.235	*11.7*	154.9	122.1	*138.5*	4.5	7.2	*5.9*
Average		33.8	8.1	*21.0*	288.2	199.6	*233.0*	13.9	22.8	*18.4*
Minimum		18.2	4.8	*11.7*	154.9	112.1	*138.5*	4.5	7.2	*5.9*
Maximum		41.9	13.2	*27.4*	372.8	387.2	*380.0*	23.4	38.3	*29.3*
LSD, P>5%				4.84			114.3			14.04

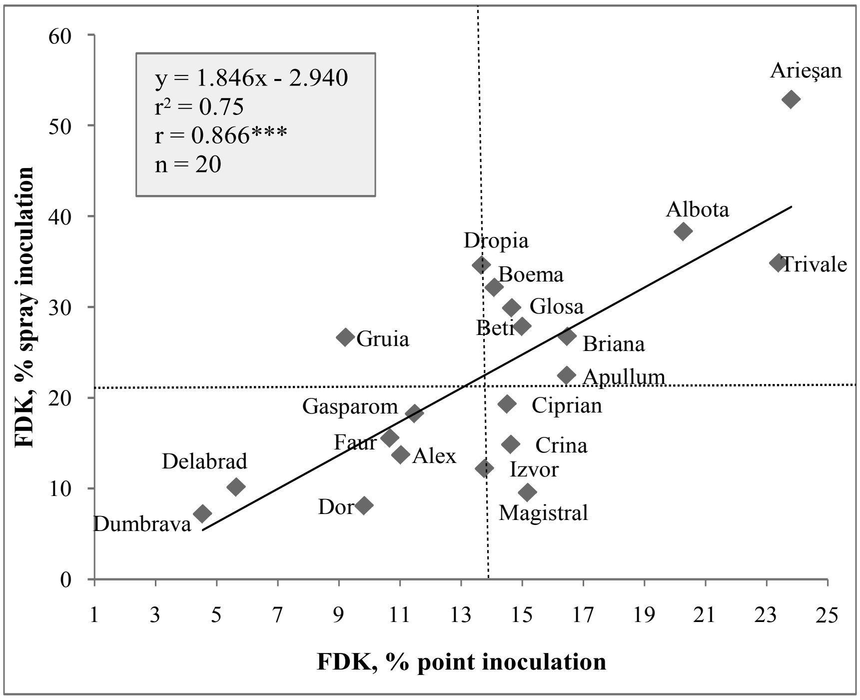

Figure 2. Correlation between point and spray field inoculation 2008-2009 (Fundulea, FDK, %, mean values)

CONCLUSIONS

In three out of 15 environments (year x location combinations) from Romania, up to 10% risks of fungus contamination under natural conditions was found, but level of DON accumulation in grain was generally under 1.25 mg/kg [(limit of contamination according to Commission Regulation (EC) No. 1126/2007]. A broad range of variation for aggressiveness vs. wheat varieties, in seedling and adult stage, has been found in *Fusarium* accessions collected from five locations. Phenotypic differences between resistance type of 20 wheat Romanian cultivars was noticed, which suggest association of resistance Type 1 and 2 (Dumbrava, Delabrad, Dor) or prevalence of Type 1 (Izvor, Crina, Magistral). Further researches are required to validate by molecular tools patterns of resistance and aggressiveness in these wheat/*Fusarium* pathosystems.

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