

Reaction of Wheat to Common Bunt and Dwarf Bunt and Reaction of Triticale to Dwarf Bunt

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Abstract

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Resistance to common bunt and to dwarf bunt in winter wheat cultivars recently registered in the Czech Republic was studied in artificially inoculated field trials in the years 2013–2015. In trials with common bunt, seeds of each experimental series were inoculated with a different mixture of isolates. In dwarf bunt trials, the soil surface was inoculated with a natural bunt population from a single locality. Several selected unregistered wheat cultivars, triticale cultivars and cultivars/lines known as sources of dwarf bunt resistance were also included in the trials with dwarf bunt. Out of the recently registered winter wheat cultivars only cv. Genius showed resistance to common bunt in both test years. Cv. Sailor was highly resistant to common bunt only in one trial, but not in other trials with different inoculum. Cv. Saturnus and the registered cv. Potenzial showed the lowest incidence of dwarf bunt in both years. The triticale cultivars were highly resistant to dwarf bunt compared to the wheat cultivars used as checks. High resistance to dwarf bunt in the tested sources of resistance was confirmed.

Keywords: bunt incidence; resistance; sources of resistance; winter wheat cultivars

Though the chemical seed treatment of wheat is effective against common bunt caused by *Tilletia caries* (D.C.) Tul. & C. Tul. (syn. *T. tritici* (Bjerk.) G. Winter and *T. laevis* J.G. Kühn (syn. *T. foetida* (Wallr.) Liro, and can also reduce dwarf bunt caused by *T. controversa* J.G. Kühn, the availability of effective host resistance is crucial for bunt control, particularly in low-input and organic farming (SPIESS 2015).

Winter wheat cultivars, registered and recommended in the Czech Republic, have been tested in Crop Research Institute for common bunt resistance each year since 1989. Our last results on common bunt resistance from the years 2009–2013 were published in 2014 (DUMALASOVÁ *et al.* 2014). This contribution presents data on cultivars newly tested for common bunt resistance from the years 2013–2015 and data on dwarf bunt resistance obtained since 2011.

MATERIAL AND METHODS

Seed of winter wheat cultivars registered at present or in the past in the Czech Republic (Table 1) originated from the Central Institute for Supervising and Testing in Agriculture, Brno, <http://www.ukzuz.cz/>. Seed

of winter wheat cultivars unregistered in the Czech Republic (Table 3) was kindly supplied by Hermann Bürstmayr, IFA Tulln, Austria. A set of differential controls consisting of lines/varieties possessing *Bt0–Bt13* genes (GOATES 1996) and the sources of resistance (Table 5) were obtained by courtesy of Dr. B.J. Goates. Seed of triticale (Table 6) was provided by the Gene Bank of the Crop Research Institute, Prague-Ruzyně, <https://grinczech.vurv.cz/gringlobal/search.aspx>.

Cultivars/lines were tested in the field at the Crop Research Institute in Prague-Ruzyně (Czech Republic) for two years as a rule. Field trials had four replicates, each of them represented by one row 1 m long, 0.2 m apart. The resistant checks Globus and Bill and the susceptible check Batis were included in the tests.

In common bunt tests, seed was inoculated with a mixture of common bunt teliospores before sowing. Inoculation was done by shaking 250 seeds with 0.1 g of teliospores in Erlenmayer flasks for 1–2 min by hand. Two different inoculum mixtures of teliospores were applied to the seed in two experimental series of two-year trials. The mixture marked “mix” was originally a mixture of six Czech isolates, prepared in 2011, reinoculated several times on susceptible

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wheat genotypes in Ruzyně. The “mix” mixture was used in the first experimental series for tests in 2013 and 2014. Another mixture, “kss”, consisting of three isolates from various Czech locations was employed in the second experimental series in the tests in 2014 and 2015. The mixtures “mix” and “kss” were tested on controls consisting of lines/varieties possessing *Bt0–Bt13* genes in the respective years of testing. In 2014 an additional mixture, marked “rukr”, was prepared with an intention to obtain wider virulence in 2015 evaluations of a restricted number of selected cultivars. Inoculations and sowing were carried out in early October. The sowing dates were 10 October 2012, 8 October 2013 and 7 October 2014.

For the purpose of race identification an infection incidence above 10% of the spikes indicates virulence (GOATES 1996). On the basis of this rule we assume that the potential for breeding for resistance of the cultivars showing more than 10% of infection already in only one-year trials is low.

The original inoculum of dwarf bunt was harvested in 2006 from our experiment on a Czech locality with natural occurrence of dwarf bunt and has been maintained by reinoculations in our plots in Ruzyně since that time. For dwarf bunt tests, eight replications were sown to the seedbeds in late October. However, only the first four out of the evaluated rows were included in the statistical analysis so that the cultivars with incomplete data sets might be taken into the analysis. One row with a susceptible winter wheat variety was inserted after every four rows to check equal distribution of the infection. Dry teliospores were evenly spread on the soil surface shortly after sowing. Soil inoculation was carried out with the doses of 2 g of teliospores per 1 m². In the absence of snow cover the plots were covered with white nonwoven fabric during winter months in order to improve conditions for infection.

Healthy and bunted ears were scored in July by counting. The reaction to bunt was expressed as a percentage of all the spikes in the row exhibiting bunt. Analysis of variance was employed to determine if statistical differences between treatment means were observed and Fisher’s Least Significant Difference (LSD) test was applied to separate means (UNISTAT 5.0 package, Unistat Ltd., London, UK).

RESULTS AND DISCUSSION

Common bunt. Average bunt incidence of all the genotypes inoculated by “mix” inoculum and included in the statistics was 60.0% in 2013 and 56.7% in 2014.

The second inoculum, “kss”, reached in 2014 the average value of bunt incidence 44.2%. In 2015 it was even less, only 28.2%. In our previous experiments the overall average values usually reached approximately 40% (DUMALASOVÁ *et al.* 2014).

Similarly, the susceptible check cultivar Batis inoculated by “mix” inoculum showed the 86.5% bunt incidence in 2013 and 84.4% in 2014, while the infection by “kss” inoculum reached 74.5% in 2014. The infection by “kss” inoculum in 2015 resulted in the bunt incidence of 43.5%. Mean levels of common bunt infection observed in cv. Batis in the previous years were around 60% (DUMALASOVÁ *et al.* 2014). The conditions in 2015 were slightly less convenient for bunt development than in other experimental years according to warmer soil temperature values from October to December recorded by an agrometeorological station, Crop Research Institute, Prague, in 2014.

The cultivar Globus has been used as a resistant check in our trials with various common bunt isolates since 2004. We suppose the presence of resistance gene/s to bunt in its genome, however, these genes have not probably been identified so far. The data presented here were obtained in the years 2013–2015. The level of incidence fluctuated between 0% and 4% of bunted spikes with no distinct differences associated with the inoculum type. Although pathotypes virulent to cv. Globus are known, the same level of resistance as in cv. Globus has not been acquired in any of the 35 genotypes included in our trials, with a single exception, cv. Genius, tested only for two years with a single inoculum mixture so far.

The resistant check cv. Bill had the bunt incidence level of 7.9% and 7.6% using the “mix” inoculum mixture in 2013 and 2014, respectively, while there was only 0.6% and 0.4% of bunted spikes when the “kss” inoculum mixture was used in 2014 and 2015, respectively.

The differences in bunt incidence on susceptible and resistant check cultivars suggested that there was a sufficient disease pressure in all years of the study.

The two inoculum mixtures used in the trials differed in virulence, as was proved in the two-year experiment on the standard set of differential genotypes performed in 2014 and 2015. The “kss” inoculum mixture has shown virulence on the lines carrying the *Bt2* and *Bt7* genes for resistance (average of the years 2014 and 2015: *Bt0* – 46.8%; *Bt1* – 6.5%; *Bt2* – 29.6%; *Bt3* – 1.0%; *Bt4* – 0.7%; *Bt5* – 5.4%; *Bt6* – 0.7%; *Bt7* – 13.6%; *Bt8* – 0.0%; *Bt9* – 0.1%; *Bt10* – 1.6%;

Bt11 – 0.0%; *Bt12* – 3.8%; *Bt13* – 1.3%). The “mix” inoculum mixture was virulent on the genotypes carrying the resistance genes *Bt2* and *Bt3* (average of the years 2014 and 2015: *Bt0* – 66.4%; *Bt1* – 0.8%; *Bt2* – 40.3%; *Bt3* – 26.6%; *Bt4* – 0.5%; *Bt5* – 1.9%; *Bt6* – 0.1%; *Bt7* – 0.0%; *Bt8* – 0.2%; *Bt9* – 0.6%; *Bt10* – 0.5%; *Bt11* – 0.0%; *Bt12* – 6.9%; *Bt13* – 1.2%).

In 2013 and 2014, fourteen registered cultivars were tested (Table 1). None of the tested genotypes reached the low infection level of resistant checks. The only exception was represented by cv. Sailor with no bunt incidence in 2014. It displayed a low infection (5.4%) and relatively low infection (22.2%) already in 2012 (DUMALASOVÁ & BARTOŠ 2013) and 2013, respectively. However, in 2015 the bunt incidence of cv. Sailor in the European *Tilletia* cooperative trial at the Crop Research Institute in Prague-Ruzyně (data not shown) reached 47.9% after infection with a new inoculum mixture “rukr”, supposed to be virulent on a wider spectrum of *Bt* genes. Cv. Sailor was further included in the European *Tilletia* Cooperative test in 2015 at many locations. It remained without bunt incidence in two trials performed in Germany (personal communication: Karl-Josef Mueller, Cereal Breeding Research Darzau, Neu Darchau, Germany; Anjana Pregitzer, LBS Dottenfelderhof e.V., Forschung & Züchtung, Bad Vilbel, Germany). At CRI in Ruzyně a different inoculum mixture employed in experiments from year to year may lead to striking differences in the results. Obviously, cv. Sailor possesses resistant gene(s) that can be overcome by a part of the Czech common bunt population. Cv. Sailor was derived from the cross (Tambor × Flair) × Drifter. Cv. Tambor, reported as resistant/medium resistant already earlier (e.g. WÄCHTER *et al.* 2007), is the probable donor of the common bunt resistance in the cv. Sailor.

Tests of 21 registered cultivars were performed in 2014 and 2015. The cv. Genius proved very high resistance in the two years of testing (bunt incidence 2.7% in 2014 and no bunt incidence in 2015). Cv. Genius originates from the same company as the earlier released bunt resistant cultivars Tommi, Globus and Quebon. According to the Recommended List of Varieties 2014 by HORÁKOVÁ *et al.* (2014), the pedigree of the cv. Genius is ACK 3094 × 00/412. It is necessary to validate reactions of the cv. Genius in additional tests using a wider set of diverse common bunt isolates to see the potential of cv. Genius for bunt control through the host resistance. Cv. Genius was followed in the bunt incidence level by cv. Etana (14.3% in 2014 and 10.9% in 2015).

Table 1. Mean levels of common bunt infection observed in field trials at Prague-Ruzyně from 2013 to 2015 (two experimental series with different sets of cultivars and inoculum)

Cultivar	Registration*	% bunted ears
Exp. 1 (2013–2014), inoculum „mix“		
Globus (check)	2003	2.9 ^a
Bill (check)	2002	7.7 ^a
Sailor	2011	11.1 ^a
Golem	2011	53.8 ^b
Dagmar	2012	56.3 ^b
Chevalier	2011	56.7 ^{bc}
Tiguan	2012	60.6 ^{bc}
Princeps	2012	63.0 ^{bcd}
KWS Ozon	2012	66.2 ^{cde}
Dulina	2013	70.9 ^{def}
Cimrmanova raná	2012	73.3 ^{ef}
Turandot	2012	73.4 ^{ef}
Hewitt	2012	76.0 ^{efg}
Citrus	2011	77.2 ^{fg}
Evina	2012	78.5 ^{fg}
Fermi	2011	79.4 ^{fg}
Batis (check)	2001	85.5 ^g
mean		58.4
Exp. 2 (2014–2015), inoculum „kss“		
Bill (check)	2003	0.5 ^a
Globus (check)	2002	0.6 ^a
Genius	2014	1.4 ^{ab}
Etana	2013	12.6 ^{abc}
Tilman	2014	14.9 ^{bc}
Nordika	2014	24.5 ^{cd}
Gordian	2014	32.4 ^{de}
Fabius	2013	35.4 ^{def}
Patras	2013	36.3 ^{defg}
Florus	2014	39.5 ^{efgh}
Zeppelin	2013	39.5 ^{efgh}
Brokat	2013	40.7 ^{efghi}
Matchball	2013	42.5 ^{efghi}
Fakir	2013	43.2 ^{efghij}
Annie	2014	44.2 ^{efghij}
Tobak	2013	45.0 ^{efghij}
Rumor	2014	45.8 ^{efghijk}
Vanessa	2013	48.1 ^{efghijk}
Avenue	2014	49.2 ^{ghijk}
Artist	2014	51.3 ^{hijk}
Julie	2014	52.5 ^{hijk}
Lavantus	2013	53.8 ^{ijk}
Tosca	2014	56.3 ^{jk}
Batis (check)	2001	59.0 ^k
Mean		36.2

Means in columns followed by the same letter are not significantly different from each other (LSD, $P < 0.05$); Exp. – experimental series; *year of registration in the Czech Republic

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Table 2. Analysis of variance for common bunt infection for the cultivars registered in the Czech Republic and evaluated in Prague-Ruzyně from 2013 to 2015

Experimental series	Inoculum	Source of variability	Sum of squares	% variation	df	Mean square	F value	P-value
1 (2013–2014)	„mix“	cultivar	86 782.43	87.92	16	5 423.90	100.69	< 0.0001
		year	364.89	0.37	1	364.89	6.77	0.0107
		cultivar × year	5 761.17	5.84	16	360.07	6.68	< 0.0001
		error	5 333.11	5.40	99	53.87		
		total	98 704.26	100.00	135	731.14		
2 (2014–2015)	„kss“	cultivar	58 578.01	64.67	23	2 546.87	32.55	< 0.0001
		year	12 291.90	13.57	1	12 291.90	157.09	< 0.0001
		cultivar × year	8 390.03	9.26	23	364.78	4.66	< 0.0001
		error	11 032.90	12.18	141	78.25		
		total	90 580.56	100.00	191	474.24		

df – degrees of freedom

Cultivars Fakir and Zeppelin, tested in the 2014–2015 experimental series, were susceptible to common bunt (average bunt incidence 43.2% and 39.5%, respectively) though they possess the resistant cultivar Tommi in their pedigrees. They originate from a different company than cv. Tommi. In both trials resistant checks had a very low bunt incidence, cv. Bill 7.7% and 0.5%, cv. Globus 2.9% and 0.6% in the 2013–2014 and 2014–2015 experimental series, respectively. The average level of bunt incidence was higher in 2013–2014 than in the 2014–2015 trials.

Due to different climatic conditions influencing the plant and fungus development in various years the variation of bunt incidence from year to year is usual even when the same inoculum mixture is used. The influence of diverse sets of cultivars and different viability or aggressiveness of the inoculum mixtures play an additional role in our trials.

Analysis of variance for common bunt incidence indicated that there was a significant effect due to cultivar ($P < 0.0001$ in both experimental series) and year ($P = 0.01$ in the 2013–2014 experimental series, $P < 0.0001$ in 2014–2015), and a significant interaction effect on cultivar-by-year ($P < 0.0001$) (Table 2). Statistically significant differences between the examined genotypes indicate a very high cultivar effect on the level of bunt infection. In the 2013–2014 experimental series the contribution of cultivars to the total variation exceeded 87%. In the 2014–2015 experimental series the cultivar effect contributed 64.67% to the total variation. Year and cultivar × year interactions were also significant, they contributed 0.37–13.57% to the total variation.

Dwarf bunt. In 2014 and 2015 fifteen winter wheat cultivars, among them cvs. Potenzial, Mulan, Batis,

Bohemia, Magister and Federer, registered in the Czech Republic at present, were tested for resistance to dwarf bunt (Table 3). The average bunt infection was 9.3% in 2014 and 5.7% in 2015. Cv. Batis, a highly susceptible cultivar to common bunt (e.g. HUBER & BURSTMAYR 2006; WÄCHTER *et al.* 2007), presumed as a susceptible check, proved only medium susceptibility to dwarf bunt in our trials, 8.9% in 2014 and 3.9% in 2015. A possible reason for the

Table 3. Mean levels of dwarf bunt infection observed in infection trials at Prague-Ruzyně from 2014 to 2015

Cultivar	Registration*	% bunted ears
Saturnus	2000 (AT)	1.71 ^a
Potenzial	2012 (CZ)	2.31 ^{ab}
Mulan	2007 (CZ)	4.55 ^{ab}
Globus	2003 (CZ)	5.28 ^{ab}
Tommi	2002 (DE)	5.52 ^{ab}
Radiant	–	5.75 ^{ab}
Batis	2001 (CZ)	6.41 ^{ab}
Bohemia	2007 (CZ)	6.48 ^{ab}
CDC Raptor	–	6.53 ^{ab}
Magister	2009 (CZ)	6.79 ^{ab}
Rainer	2006 (AT)	7.12 ^{ab}
Federer	2009 (CZ)	7.26 ^b
Capo	2000 (HU)	14.12 ^c
Midas	2008 (AT)	14.71 ^c
Pannonikus	2008 (AT)	17.66 ^c
Mean		7.48

Means in columns followed by the same letter are not significantly different from each other (LSD, $P < 0.05$); *year of registration in the Czech Republic or other EU country

Table 4. Analysis of variance for dwarf bunt infection for the cultivars evaluated in Prague-Ruzyně from 2014 to 2015

Source of variability	Sum of squares	% variation	df	Mean square	F value	P-value
Cultivar	2 271.60	41.44	14	162.26	6.24	< 0.0001
Year	394.93	7.21	1	394.93	15.19	0.0002
Cultivar × year	522.87	9.54	14	37.35	1.44	0.1536
Error	2 261.43	41.26	87	25.99		
Total	5 481.32	100.00	119	46.06		

df – degrees of freedom

Table 5. Dwarf bunt incidence values in the infection tests of sources of resistance conducted at Prague-Ruzyně from 2013 to 2015

No incidence	702_1102C (2015), Blizzard (2013, 2014, 2015), Bonneville (2015), Franklin (2013), PI119333 (2014), Golden Spike (2013, 2014, 2015), M82-2123 (2013, 2014, 2015), PI178383 (2013, 2014, 2015), PI560603 sel blaw (2013), PI560795 sel bcors (2013), PI560841 sel bcl (2014), Thule III (2013), Weston (2015)
0.1%	702_1102C (2014), Bonneville (2013, 2014), PI119333 (2015), Weston (2013)
0.2%	PI560841 sel bcl (2013, 2015)
Batis (susceptible check)	2013 – 4.2%; 2014 – 8.9%; 2015 – 3.9%

low disease level in the 2015 trial may be the absence of long-term snow cover demanded for the optimal development of dwarf bunt infection. Though the plots were covered with white unwoven fabric to improve the infection level, it exceeded the 10% level only in several cases. Despite the fact that the reactions may be not fully demonstrated, the genotypes showed a consistent performance over years with no considerable differences in the infection level with the significant Pearson correlation coefficient of 0.51 (Figure 1). Analysis of variance for dwarf bunt incidence indicated a significant effect due to cultivar ($P < 0.0001$) and year ($P < 0.0002$) (Table 4).

The group with the highest dwarf bunt incidence comprised three cultivars. Cvs. Capo, Midas and Pannonikus clearly expressed susceptible reactions (Table 3). Cv. Globus is known to possess resistance to some races of common bunt. Surprisingly, the level of dwarf bunt infection on closely related cvs. Globus and Tommi (DUMALASOVÁ *et al.* 2014) was not very distant from the value reached in cv. Batis. Cv. Globus showed a lower susceptibility to dwarf bunt in our large-scale field trials. Increased levels of infection with dwarf bunt on cv. Globus were reported by e.g. HUBER and BUERSTMAYR (2006) from Austria.

The lowest dwarf bunt incidence (1.71%) was recorded in the cv. Saturnus. However, BAUER *et al.* (2015) reported a similarly increased level of infection in cvs. Globus, Saturnus and Tommi. The second lowest dwarf bunt infection was obtained in cv. Potenzial (2.31%), despite the fact that it is supposed

to be susceptible to the prevalent races of common bunt (DUMALASOVÁ *et al.* 2014) with the average common bunt incidence of 42.6% in 2011–2013.

Cv. Potenzial fell into a group of ten cultivars without statistically significant difference in bunt incidence. The practical significance of differences between these cultivars is also low, as the limits for the incidence of bunt spores in seed are very strict and a zero bunt incidence is sought for the effective bunt control.

Sources of dwarf bunt resistance were tested in 2013, 2014 and 2015 (Table 5). The majority of the tested genotypes remained without dwarf bunt incidence. Cvs. Bonneville, Weston and lines 702_1102C and PI119333 displayed a very low dwarf bunt incidence, similarly the line PI560841 sel bcl (0.2% in 2015).

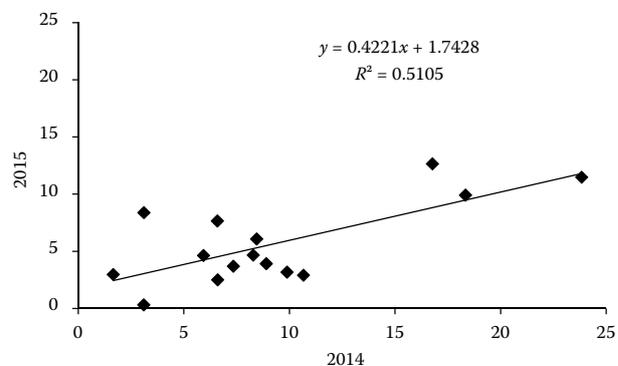


Figure 1. Relationship between dwarf bunt incidences in 2014 and 2015 (in %) for fifteen cultivars

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Table 6. Dwarf bunt infection levels in triticale in % of bunted ears

Cultivar	Registration*	2011	2012	2013	2014
Triticale					
Agrano	2008 (CZ)	6.7	0.0	3.3	0.3
Inpetto	2007 (CZ)	0.7	0.0	–	–
Dinaro	2005 (NL)	0.3	0.0	–	–
SW Talentro	2007 (CZ)	0.0	0.0	–	–
Nazaret	2005 (SK)	0.3	0.0	–	–
Hortenso	2008 (CZ)	0.1	0.0	–	–
Wheat					
Batis	2001 (CZ)	–	0.6	4.2	8.9

*year of registration in the Czech Republic or other EU country

In addition to wheat cultivars, six winter triticale cultivars were also tested for dwarf bunt resistance (Table 6). Four of them are registered in the Czech Republic at present: Agrano, Inpetto, Hortenso and SW Talentro. Dwarf bunt incidence in all the tested cultivars was very low. It did not exceed 1% in most of the genotypes. An exception was cv. Agrano with 6.7% and 3.3% bunt incidence in 2011 and 2013, respectively.

Cv. SW Talentro was free of bunt also in our previous experiments with seventeen cultivars tested in 2006 and 2008 for common bunt resistance. Common bunt incidence in the other cultivars was very low, with no bunt incidence in most of them.

Our recent trials confirmed a high common bunt resistance of cvs. Bill and Globus to most races of common bunt recorded in our previous papers. Various reactions of the cv. Sailor in different years suggest the presence of virulence in the Czech common bunt population to the relevant resistance gene(s) in cv. Sailor. Cv. Genius proved to be a novel common bunt resistant cultivar identified among winter wheat cultivars recently registered in the Czech Republic.

Cvs. Saturnus and Potenzial proved the lowest bunt incidence in the tests with artificial dwarf bunt infection.

Resistance of triticale to common bunt in the Czech Republic was already described in our previous work. High resistance also to dwarf bunt was recorded in the 2011–2015 trials comprising six winter triticale cultivars. This leads to a suggestion that in the soils infected with dwarf bunt a replacement of wheat by resistant triticale cultivars may reduce losses caused by dwarf bunt.

The tested sources of resistance proved high resistance to dwarf bunt. As they were recorded resistant to common bunt as well, they offer a suitable genetic material for resistance breeding both to common and dwarf bunt. Some of them were already used effectively in wheat breeding in North America.

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