Agronomy Research **8** (Special Issue III), 653–662, 2010

Take-all resistance of Lithuanian winter wheat breeding lines

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Abstract. Take-all, caused by *Gaeumannomyces graminis* var. *tritici*, is one of the most important root diseases of wheat around the world. Previous research has suggested that winter wheat varieties pose no effective resistance. The experiment was carried out in the winter wheat mono-crop nursery during 2006–2009. In total, 324 accessions, including standard cultivars, were tested during 3 vegetation seasons. The accessions were grown in 2.0 m^2 plots. The disease severity was assessed as ear discoloration symptoms from early to late milk stages in scores, using the scale 1 to 9 points, where 1 is the lowest value. Disease severities were high during all three seasons and ear symptoms developed from just visible to full discoloration during 3 to 5 weeks depending on the accessions' resistance and year. Take-all severity on the accessions tested was estimated from 4.75 to 9.0, 5.38 to 8.95, and 4.00 to 8.53 points in 2007–2009, respectively. Varieties 'Flair' and 'Dream' were the most frequent in the pedigree of the most resistant lines, occurring in 23.3% pedigrees of the lines. The thousand kernels and hectolitre weight showed no or low correlation with disease severities. Lines resistance showed weak correlation with yield when all plot data were used for calculation. However, correlation coefficients considerably increased when ten percent of each minimal, mean and maximal yields values were used. A mean yield of resistant and susceptible lines differed about 1 t ha^{-1} in 2007 and 2008, whereas a lower difference (0.54 t ha^{-1}) was found in 2009. Some susceptible lines had higher or similar yield as well as the resistant ones in all years.

Key words: *Gaeumannomyces graminis*, winter wheat, resistance

INTRODUCTION

The area devoted to winter wheat has considerably increased due to its larger proportion in crop rotations during recent decades. However, this cultural shift negatively affects the yield of subsequent wheat crops. One of the important reasons for yield reduction is the root rot disease "Take-all" (*Gaeumannomyces graminis* var. *tritici*) (Sieling et al., 2007). All yield components (plant, ear and grain number per m^2, grain number per ear and 1000 kernel weight) can be affected (Hornby et al., 1998). High temperatures and humidity in early autumn are known to favour plant and fungal mycelium growth, thereby increasing the probability of contact and infection (Lucas et al., 1998). The weather events' overview and general climate change forecasts in wide and narrow regional perspective (Lavalle et al., 2009; Romanovskaja et al., 2009) suggests climate warming. This trend seems to be favourable for further increasing the harmfulness of Take-all in Northern and Western Europe.

Non-regarding crop rotations (Ennaïfar et al., 2007) and management techniques (Hiddink et al., 2005), other means are available for reduction of yield losses. Factors such as sowing date, seed rate, and fertilizer strategy can have an effect on management of the disease (Cromey et al., 2006; Sieling et al., 2007). Seed treatment has limited effect due to few effective and economical fungicides in some years and sites (Dawson & Bateman, 2001; Sieling et al., 2007). Experiments with biological control of Take-all showed promising results (Zafari et al., 2008) due to a similar mechanism of pathogen suppression by other microorganisms occurring in monoculture systems of winter wheat fields (Lebreton et al., 2004).

Generally, the most efficient mean for disease control is flexible use of plant resistance. Over the several last decades, comprehensive and highly sophisticated researches have been completed concerning this disease, but clear and considerable differences among a sizeable number of winter wheat genotypes were not reported (Cook, 2003; Ennaïfar et al., 2007; Kwak et al., 2009). Some previous investigations showed differences among genotypes under greenhouse tests, but these results were not confirmed under field conditions (Penrose, 1994). Still, it remains that some wheat cultivars perform better than other in the presence of Take-all (Wallwork, 1989; Penrose, 1995; Bailey et al., 2006). Information about resistance of European winter wheat cultivars is even less available. Several decades of intensive researches showed doubtful results regarding differences of winter wheat cultivars' resistance based on root reaction evaluations. Therefore, this paper describes research on reactions of winter wheat genotypes resistance based on aboveground plant parts.

MATERIALS AND METHODS

The experiment was carried out in the winter wheat mono-crop nursery at the Institute of Agriculture (55°23′N, 23°51′E) during the period 2006–2009. The soil of the experimental site was light loamy *gleyic cambisol*, containing 2.5–2.6% humus, available phosphorus (P_2O_5) ranging from 190 to 240 mg kg^{-1}, available potassium (K_2O) from 180 to 260 mg kg^{-1} and pH between 7.0 and 7.2.

The wheat mono-crop nursery was established in 2005. Spring wheat was sown at the end of April, covering the entire nursery. The crop was chopped at the beginning of August and incorporated into the soil by disc harrow. Winter wheat breeding lines were sown in autumn 2005 under conditions described below. However, heavy drought in 2006 did not reveal considerable differences among the genotypes tested and the results were not included for analysis.

Field plots were sown at a rate of 450 kernels m^{-2} during the first decade of September. The seeds were chemically non-treated. Ploughing was done at two weeks and the seedbed was prepared one day before sowing. Fertilizer rate in pure elements (NPK) 30–60–90 kg ha^{-1} was applied before sowing and nitrogen (ammonium nitrate) 90 kg ha^{-1} was applied after resumption of vegetation. The accessions were grown in 2.0 m^2 plots in four replications without growth retardants. Weeds were controlled by herbicide use in autumn. No other pesticides were applied during plant vegetation. The material subjected to Take-all resistance tests included advanced breeding lines developed basically using European winter wheat cultivars. In total, 324 accessions, including standard cultivars, were tested during 3 vegetation seasons. A total of 137 accessions were evaluated in 2007, 117 in 2008, and 70 in 2009. The disease severity (DS) was assessed as ear discoloration symptoms from early to late milk development stages in scores, using the scale 1 to 9 points, where 1 is the lowest value. This period lasted 3–4 weeks depending on the year. DS scoring was done 3 times, approximately once a week. Take-all agent *Gaeumannomyces graminis* was dominant among fungi isolated from infected roots of winter wheat collected in the experimental nursery. Isolation and identification was done according to Hornby et al. (1998).

The correlation between DS assessments and grain yield, 1000 kernel weight, and hectolitre weight were counted as Pearson's correlation coefficients, which were evaluated for significance at probability levels 0.05 and 0.01.

The calculation of correlations between DS and hectolitre weight (HW), 1000 kernels weight (TKW), and yield consisted of three sets of plots sorted by yield. The first, correlation coefficients, were calculated between all plots, according to the mentioned above traits. The second counting involved each 10% of plots with minimal, mean and maximal values each (sorting A). The third calculation involved each 10% of plots with minimal and maximal values each (sorting B). Selected lines DS and yield values were evaluated for significance at probability level 0.05.

Weather conditions were favourable for high grain yield formation due to non-stressing winters and sufficient precipitation during vegetation periods. Disease development was also positively influenced by long warm autumns and short warm winters.

RESULTS AND DISCUSSION

The experimental seasons slightly differed in terms of conduciveness to the development of Take-all. DS was high during all the three seasons and ear symptoms developed from just visible to full discoloration during 3 to 4 weeks depending on the accessions' resistance and year. DS was evaluated 3 times during 2007 and 2009, and 4 times in 2008. However, the first evaluation in 2009 was not considered due to low DS. Fig. 1 shows the response of winter wheat to disease. The highest differences among the accessions were identified in 2007.

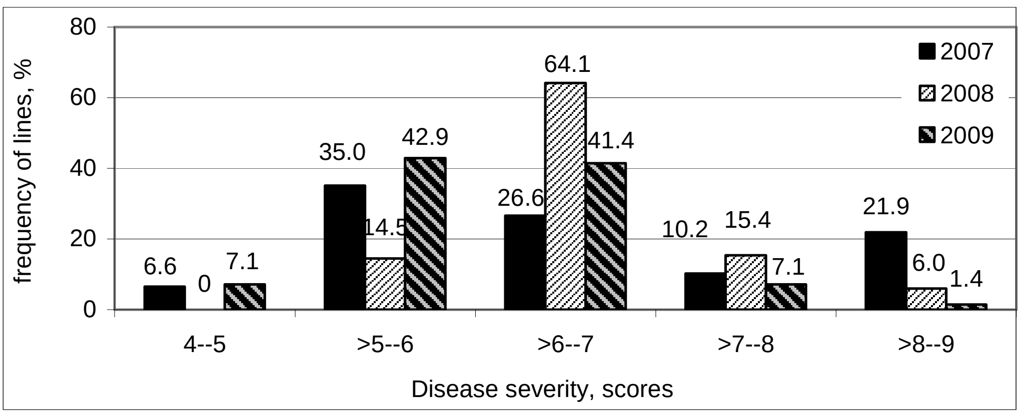

Figure 1. Distribution of winter wheat accessions by resistance to Take-all in 2007–2009.

Take-all severity on the accessions tested was estimated from 4.75 to 9.0 points in 2007. The genotypes diseased 4–5 and 5–6 points accounted for 6.6 and 35.0% of those tested. Severely infected (8–9 points) genotypes were rather frequent (21.9%). The accessions were characterized by points from 5.38 to 8.95 in 2008. The genotypes evaluated by 6–7 points dominated. The results of this year differentiated genotypes slightly insufficiently. However, about 20% of genotypes could be eliminated due to a high disease level compared with the rest (Fig. 1). The infection of accessions in 2009 was adequate for elimination of susceptible genotypes as lines were infected from scores of 4.00 to 8.53 and accessions infected over 6 points amounted to about 50%. The standard varieties (‘Zentos’ and ‘Ada’) showed quite stable resistance level across the 3-year period indicating relatively stable disease pressure on the genotypes tested.

Table 1. The correlation coefficients among Take-all severities and yield, thousand kernels weight and hectolitre weight.

Take-all evaluations	Trait	All plots yield values	Plots with min 10% + mean 10% + max 10% yield values	Plots with min 10% + max 10% yield values
		2007		
1		-0.219	-0.371	-0.492*
2	Yield	-0.486*	-0.599*	-0.683**
3		-0.351*	-0.449*	-0.548*
1		-0.429*	-0.593**	-0.639**
2	TKW†	-0.535*	-0.707**	-0.789**
3		-0.389	-0.535**	-0.545**
1		-0.248	-0.305*	-0.376*
2	HW††	-0.339*	-0.433*	-0.423*
3		-0.293	-0.219	-0.208
		2008		
1		-0.490*	-0.667**	-0.605**
2	Yield	-0.216	-0.732**	-0.602**
3		-0.227	-0.335*	-0.285
1		-0.103	-0.210	-0.139
2	TKW	-0.077	-0.266	-0.275
3		0.003	-0.032	-0.047
1		0.059	-0.113	-0.072
2	HW	0.126	-0.013	-0.016
3		0.219	0.053	0.068
		2009		
1		-0.564*	-0.715**	-0.734**
2	Yield	-0.081	-0.086	-0.103
3		-0.059	-0.039	-0.047
1		-0.249	-0.509*	-0.601**
2	TKW	0.077	0.139	0.087
3		0.089	0.148	0.177
1		-0.243	-0.356	-0.354*
2	HW	-0.036	0.188	0.188
3		-0.100	0.175	0.198

†TKW – thousand kernels weight, †† HL – hectolitre weight, *, ** probability level 0.05 and 0.01, respectively

The use for calculation of correlation between DS and HW for all plots' data revealed weak negative (r = –0.339*) correlation in 2007 in the case of the 2nd DS, and no correlation in 2008 and 2009 (Table 1).

Analysis of sorted data showed slightly weak negative correlations (r = –0.305* – –0.433*) with the 1st and 2nd DS, in both cases. Only weak negative correlation was counted with the 1st DS in 2008.

The weak negative non-significant correlations were obtained among the 1st and 3rd DS and TKW and medium correlation was found between the 2nd DS and TKW in 2007. Results of the next year showed no correlations using all plot data. Data sorting considerably increased the correlation level between DS and TKW. The medium negative correlations were obtained among 1st and 3rd DS and TKW. Strong correlation was found between the 2nd DS and TKW in 2007. Data for 2008 showed no correlation between DS and TKW after sorting. The medium negative correlations were counted after sorting only for the 1st DS and TKW in 2009.

The relationship of yield with DS was low because weak negative correlations (r = –0.351* – –0.490*) were counted in 2007 and 2008; medium negative correlation (r = –0.564*) was counted in 2009 only for the first DS assessment when all plot data were used. The use of sorted and selected data increased correlations. The sorting A increased negative Take-all impact on yield as weak to medium negative (r = –0.371, not significant – –0.599*) correlations were found for all DS in 2007. The sorting B increased the relationship more as correlations increased (r = –0.492* – –0.683**) in 2008. Data sorting showed considerable increase of correlation in 2008. The sorting A showed weak to strong negative (r = –0.335* – –0.732**) correlations. The sorting B showed medium negative correlation (r = –0.602** – –0.605**) for the 1st and 2nd DS. The strong negative (r = –0.715** – –0.734**) correlations were counted in the case of both sorting for the 1st DS in 2009. The 2nd and 3rd DS showed no correlations.

The breeding lines presented in Table 2 were the most susceptible and the most resistant among the genotypes of the screening years. The differences among the genotypes were significant for resistance to Take-all if comparing susceptible lines with resistant. The least damaged lines were evaluated by scores of 5.0–5.2 in 2007. The most susceptible lines were characterized by scores of 8.5–8.9. The difference in Take-all reaction between the most distinct lines was 1.8 times. The most resistant lines had scores from 5.4 to6.0 and the most susceptible ones by scores of 7.3–8.1 in 2008. The most resistant lines were evaluated by scores from 4.0 to 5.4 and the most susceptible from 6.6 to 8.5 in 2009. The varieties 'Flair' and 'Dream' were the most frequent in pedigree of the most resistant lines. They occurred in 23.3% of the lines, whereas only 6.7% of the most susceptible lines possessed these varieties. The frequency of the remaining varieties in the pedigrees of resistant lines was too low for definition of their impact. About half the susceptible lines possessed geographically fewer related varieties from the continental climate countries. 'Rostovchanka' was one that occurred in the pedigree of 13.3% of the lines.

The yield mean of resistant and susceptible lines differed by about 1 t ha^{-1} in 2007 and 2008, whereas a lower difference (0.54 t ha^{-1}) was found in 2009. Some susceptible lines had higher or similar yield as well as the resistant in all years. The highest differences were found in 2007 when only one line 'Biscay/Pesma' possessed a similar yield to many resistant ones. About half the lines had similar yields in 2008 and 2009.

The present study revealed considerable differences in Take-all resistance among the recently developed Lithuanian winter wheat breeding material.

Table 2. The winter wheat breeding lines most and least resistant to Take-all.

The most resistant lines	*DS	Yield, t ha^{-1}	The most susceptible lines	DS	Yield, t ha^{-1}
2007					
‘Flair/Pentium’	5.0	6.18	‘Elena/Lut.956’	8.5	4.53
‘Flair/Lut 9-392’	5.0	6.15	‘Rostovchanka/Flair’	8.6	3.97
‘Maverich/Lut 9-321’	5.0	5.23	‘Miron.61/WTPT128WM’	8.6	4.33
‘Miron.32/Soissons’	5.1	5.12	‘Belisar/Rufa’	8.6	3.60
‘Lone/Inna//Lut 96-2’	5.1	5.38	‘Rostovchanka/Ebi’	8.7	4.60
‘Dream/Pesma’	5.2	5.03	‘Biscay/Pesma’	8.8	5.37
‘Astron/Manef’	5.2	5.58	‘Astron/Tarso//Ukr.Od.’	8.8	3.82
‘Lut 96-3/Bold’	5.2	5.03	‘Rostovchanka/Lut.96-3’	8.8	4.52
‘Maverich/Savannah’	5.2	5.10	‘Zolotava/Miron.Ost. ’	8.9	3.12
‘Flair/Ansgar’	5.2	5.60	‘Dream/Pesma’	8.9	3.47
Mean	5.1	5.44		8.7	4.21
**$LSD_{0,05}$	0.48	1.37		0.40	1.31
2008					
‘Dream/Flair’	5.4	8.08	‘Pegassos/Belisar	7.3	7.79
‘Olivin/Anthus’	5.6	9.18	‘Belisar/Rufa	7.5	6.64
‘Dream/91002G2.1’	5.6	8.92	‘Rostovchanka/Belisar’	7.6	6.46
‘Bill/Dream’	5.6	8.22	‘Pegassos/Biscay’	7.6	8.64
‘Biscay/Dream’	5.8	9.02	‘Flair/Pentium’	7.6	8.49
‘Biscay/Flair ’	6.0	8.94	‘WW2498/Corvus’	7.9	7.83
‘Pegassos/Residence’	6.0	9.50	‘Marabu/Ansgar’	8.0	8.10
‘Lut.9329/Solist’	6.0	9.76	‘Dream/Convent’	8.0	8.29
‘Dirigent/Cortez’	6.0	8.81	‘Maverich/Savannah’	8.0	8.57
‘Flair/Haldor’	6.0	9.93	‘STH1096/96-101’	8.1	8.33
Mean	5.8	9.04		7.8	7.91
$LSD_{0,05}$	0.38	0.88		0.40	1.32
2009					
‘Astron/Bill’	4.0	6.43	‘MV Emma/Convent’	6.6	5.84
‘Astron/Olivin’	4.6	7.02	‘MV 0695/Aspirant’	6.8	4.95
‘Bill/Dream’	4.6	6.09	‘Tarso/Lut.96-3’	6.9	5.18
‘Lut.9329/Solist’	5.1	6.51	‘SW Harnesk/Olivin’	6.9	6.32
‘Lut.9329/Kornett’	5.1	6.90	‘Aspirant/Revelj’	7.0	6.57
‘Haven/Dean//Pentium /3/Cortez’	5.1	6.93	‘Lut.9392/Brandt’	7.1	6.66
‘Dream/Aspirant’	5.3	7.11	‘Biscay/Pobeda’	7.3	5.70
‘Marshal/Samyl’	5.5	5.83	‘MV 0695/Dekan’	7.3	6.33
‘Flair/Ansgar’	5.5	6.31	‘Residence/Cubus’	7.3	6.00
‘Olivin/Cubus’	5.6	6.29	‘Zentos/LIA4930’	8.5	6.46
Mean	5.4	6.54		7.2	6.00
$LSD_{0,05}$	0.69	0.99		0.52	0.88

*DS – maximal diseases severity, **$LSD_{0,05}$ – least significant difference at probability 0.05

It is good news for winter wheat breeders, as previous reports on resistance screening did not reveal such considerable differences (Wallwork, 1989; Penrose, 1995; Hornby et al., 1998; Bailey et al., 2006). Previous resistance screenings were based mainly on root rot level. It seems that evaluation of ear discoloration characterized genotypes by resistance would be more presentable. This method should be more precise because of highly specific aboveground plant reaction to root rot caused by *G. graminis*. Also it was plant non-destructing and much faster when compared with root evaluation for genotypes' evaluation speed, which is highly important for screening of early breeding material grown by the thousands of small plots or headlines. Only one of a few studies suggested the use of ear symptoms for Take-all evaluation (Kabbage & Bockus, 2002).

There were often large differences within a line in different replications. Such differences were probably due to the uneven distribution of soil-borne inoculum within the trial. The high level of Take-all in some lines indicated that they had the potential to be heavily infected. Conversely, those lines that had low disease levels in all years can be reasonable claimed to be resistant to this disease, although confirmation in further trials with controllable infection levels and over wider locations would be desirable. Slightly different weather conditions among years were favourable for discrimination of quantitative resistance level. It is likely that resistance of the material tested depended entirely on this resistance type, as there were no lines possessing high resistance and reaction to disease distributed closely to normal.

Lower disease incidence in 2009 could be related to the decline of *G. graminis* population in soil due to the spread of natural antagonists (Simon & Sivasithamparam, 1989; Lebreton et al., 2004; Zafari et al., 2008). Lower disease reactions showed the possibility to discriminate more genotypes with satisfactory resistance level. *G. graminis* is a necrotrophic fungus (Hornby et al., 1998; Kabbage & Bockus, 2002); therefore inoculum in the screening site should not be too concentrated, to avoid destruction and resistant accessions.

One explanation for differences in resistance of varieties is the ability of the differing winter wheat genotypes to use manganese. Manganese might increase the biosynthesis of defence-related phenolic and lignin and thus resistance to Take-all (Wilhelm et al., 1987; Rengel et al., 1993). The level of this element is from sufficient to high according to data of its distribution in Lithuanian soils. Therefore, only a very small share of breeding lines could be more affected by Take-all due to the manganese level.

Penrose (1987) suggested that wheat had two resistance levels, which although low, could be useful. Differences in the thickening of cortical cell walls in the seminal roots of wheat seedlings, providing a mechanical barrier to infection, seemed to be responsible for differences in the susceptibility of cultivars to Take-all.

The use of sorted data for calculation of correlations showed a tendency that the more genotypes differ by resistance reaction, the higher relationship between yield and disease severity exists. It is one of the main reasons why using numerous breeding lines relation for calculation is low contrary to results of experiments with selected genotypes and infection level (Kabbage & Bockus, 2002; Ennaïfar et al., 2007; Sieling et al., 2007). However, data sorting did not considerably increase the correlation between HW, TKW and Take-all severities. Possibly, small plots used for screening markedly affected yield and its elements due to the strong effect of outside rows.

Generally, it was stated in previous researches that winter wheat possesses low resistance to Take-all. The use of wild relatives of related species such as rye and triticale is suggested for resistance breeding. Also, there is a possibility of transferring the ability to produce avenacin, thought to be a cause of resistance to *G. graminis*, from oats to wheat (Conner et al., 1988; Eastwood et al., 1993; Liu et al., 2001; Cox et al., 2005). However, up to now no publications about efficient transfer of resistance from wild relatives is available. Nevertheless, our results show that a considerable resistance level was achieved when widely available cultivars were used for breeding.

CONCLUSIONS

Disease severities were high during all the three seasons and ear symptoms developed from just visible to full discoloration during 3 to 4 weeks depending on the accessions' resistance and year. Take-all severity on the accessions tested was estimated from 4.75 to 9.0, 5.38 to 8.95, and 4.00 to 8.53 scores in 2007–2009, respectively. The varieties 'Flair' and 'Dream' were the most frequent in pedigree of the most resistant lines. The frequency of the other varieties in pedigrees of resistant lines was too low for definition of their impact. About half the susceptible lines possessed geographically fewer related varieties from continental climate countries. Generally, thousand kernel and hectolitre weight showed no or low correlation coefficients using all plots data for calculation as well as 10% of each minimal mean and maximal values sorted by yield. Lines resistance showed weak to medium correlation with disease severities when all plot data were used. However, correlations considerably increased to medium-strong when calculations were made with sorted data selecting 10% of each minimal mean and maximal yields values. The yield mean of resistant and susceptible lines differed by about 1 t ha^{-1} in 2007 and 2008, whereas lower difference (0.54 t ha^{-1}) was found in 2009. Some susceptible lines had higher or similar yield as well as the resistant ones in all years.

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