

Sprouting resistance and Falling Number values in introgressive *Triticale*/*T. monococcum* lines

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Abstract

The aim of this work was to evaluate how the Falling Number (FN) in introgressive triticale lines is influenced by introduced *Triticum monococcum* genes responsive for mature spikes sprouting resistance (SR). These lines were developed in two independent series (A and B) by incorporating of diploid wheat (*T. monococcum*) genes in two different strains of hexaploid triticale (LT 176/10 and LT 522/6) using the synthetic allotetraploid *T. monococcum*/*Secale cereale* ($A^m A^m RR$) as a bridging form. In three consecutive vegetation seasons the mature spikes response to artificial wetting was tested to assess SR. FN was evaluated in full milling of seeds to characterize the level of starch-protein matrix damage in 57 introgressive triticale lines. The obtained results showed twofold higher coefficients of variation of the SR than FN. SR-FN correlation was generally significant in a B-series of lines and statistically insignificant in an A-series showing dependence on the triticale recipient parent and year. In opposite to SR, in both series there was not a line having FN comparable with that of diploid wheat parent. This demonstrates that genetic background of the FN is more complicated in relation to inheritance of SR.

Additional key words: artificial wetting, endosperm damage, variation coefficients, year effect.

Introduction

The characteristic feature of triticale seeds physiology is their high sensitivity to sprouting (McEvan and Haslemore 1983, Derera 1989, Doliński 1995). In years abounding with rains at harvesting time the kernels germination in the spikes diminishes the seeds values as a reproductive material. Sprouting sensitivity is also

Received 21 January 1999, accepted 2 June 1999.

Acknowledgement: This work was supported by a grant No. 60/HiE/5K-118/98 from the Agricultural Property Agency of State Treasury - Warsaw Branch, Poland.

The paper was presented on the conference "8th Days of Plant Physiology" held in Olomouc, Czech Republic, 7-10 July 1998.

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one of the barriers stopping the efforts to develop the lines good enough in milling and backing properties.

Spike sprouting is influenced by some of spike characters and by a chain of physiological events that lead up to *de novo* synthesis of hydrolytic enzymes, which causes the endosperm changes - first of all the starch decompose by α -amylase activity (Mares 1989). The synthesis of isozymes of α -amylase which are normally associated with the early stages of germination is controlled by the α -Amyl genes located on the group 6 of wheat chromosomes. In ripening grains of some cultivars α -Amyl genes produce high levels of enzyme even in the absence of rain (Mares and Gale 1990). This phenomenon has been termed "late maturity α -amylase".

In triticale, the chromosome arm 6RL was shown to control *de novo* synthesis of α -amylase (Gale *et al.* 1990), and this arm was found to be responsible for increased enzyme synthesis during late stages of triticale grain maturation (Masojć *et al.* 1996). Induction of α -amylase synthesis in maturing grain usually results in reduced quality of flour (Mares and Miva 1993). The level of starch decay evaluated by viscometric enzyme activity assay as Falling Number (FN) value is still one of the commonly used method of determining the kernel damages caused by α -amylase activity (Dyck *et al.* 1986, Mares 1989, Upadhyay and Paulsen 1988, De Pauw and McCaig 1991, Trethowan 1995).

In case of the hexaploid wheat, the damaging of starch-protein matrix of seed endosperm showed by diminishing FN could be significantly restricted by the introduction of genes responsive for pre-harvest sprouting resistance (Czarnecki 1987, De Pauw *et al.* 1982, 1985). Expression of these genes was associated with red colour of seed coat and/or seed dormancy character (De Pauw and McCaig 1984, Dyck *et al.* 1986, Flintham 1993). Although structural genes encoding α -amylase synthesis were identified, little is known about regulatory genes which affect the enzyme content in cereal grains. Masojć *et al.* (1996) reported that in crosses between rye lines low enzyme activity showed complete dominance over high level of its synthesis. Segregation ratios indicated that two independent duplicative loci underlie high or low α -amylase production.

The introduction of *Triticum monococcum* genes into hexaploid triticale resulted in sprouting resistance of introgressive lines with frequency showing dominance of this character (Sodkiewicz and Tomczak 1994). The aim of this work was to evaluate how influenced are the Falling Number values in introgressive triticale lines with introduced *Triticum monococcum* genes responsive for mature spikes sprouting resistance.

Materials and methods

The studying material consisted of two independent series (A and B) of introgressive triticale lines (Tc/Tm) developed as a result of introducing A^m - genome of *Triticum monococcum* L. var. *macedonicum* Papag. in two strains of hexaploid triticale (LT 176/10 - series A; LT 522/6 - series B). This *T. monococcum* variety in earlier

studies distinguished by a markedly noticeable tolerance to spike wetting (Sodkiewicz and Tomczak 1994). Synthetic allotetraploid *T. monococcum* var. *macedonicum* / *S. cereale* cv. Dańkowskie Żłote ($A^m A^m RR$) was used as a "bridging form" in crosses with forementioned triticales. All introgressive Tc/Tm lines were obtained from *Triticale* \times $A^m A^m RR$ recombinants as a secondary hexaploid progeny resulting from chromosome number increase in progenies plants of BC₁ with parental triticales (Sodkiewicz 1992).

A^m/A genomes recombination took place until BC₁/F₄ generation resulting in introducing some parts of unknown amount of genetic information from *T. monococcum* into secondary hexaploids, which was evident from observed segregation of *T. monococcum* characters, among them the sprouting resistance (Sodkiewicz 1992, 1994, 1996). The developed morphologically uniform, introgressive Tc/Tm lines (BC₁/F₅, BC₁/F₆, BC₁/F₇ generations) and the parental genotypes were grown in three consecutive seasons (experiment I, II, III) at the experimental field of the Institute of Plant Genetics PAS in Cerekwica (about 30 km from Poznań). Samples of 15 spikes at full physiological maturity were collected from each of 57 Tc/Tm line as well as their parental strains. In every season the mature spikes response to artificial wetting was tested to assess sprouting resistance (SR) and Falling Number (FN) was evaluated in full milling of seeds to characterize the level of starch-protein matrix damage.

SR was assessed by artificial wetting of intact spikes in a germination blotter test according to Hagemann and Cihra (1984) and expressed as an average number of germinated seeds per spike. Testing was conducted at relative seed humidity lower than 12 %. Falling Number (FN) values (according to Perten-Hagberg) were evaluated for ~30 g of mill, in two repetitions with 7 g of mill, corrected according to relative water content, using Falling Number apparatus type 1800 (*Perten Instruments Co.*, Huddinge, Sweden). Preparation of samples tested was done following ISO-Standard No. 3093 and ICC Standard No. 107.

Linear correlations coefficients SR-FN were calculated, and linearity were proved with variation analysis in linear regression.

Results and discussion

Among parental strains which genetic material was transmitted into introgressive lines, the diploid wheat showed the highest level of SR and parallelly its FN value in average of three years was the highest (Table 1). Hexaploid triticales (a recipient parents) differed in their tendency to sprouting as well as in FN. LT 522/6 triticales in both characters was better and much close to rye Dańkowskie Żłote and *T. monococcum* - the donors of genes in introgression process. Generally, in group of parental strains SR of mature spikes assessed as an average number of sprouted kernels per spike showed no statistically significant correlation with FN (Table 1).

The introgressive triticales lines (Tc/Tm) showed high variability of both analyzed characters. The spikes sprouting variability expressed as coefficient of variation (CV) was about twofold higher in relation to the Falling Number variability (Table 2). In

Table 1. Spike sprouting resistance (SR) and kernels Falling Number (FN) averaged over 3-years in parental strains and introgressive triticale hexaploid lines. Correlation coefficient between SR and FN $r = -0.781$.

Parents	SR	FN	Lines	SR	FN
<i>T. monococcum</i>	1.4	372.0	LT 176/10	29.9	71.3
<i>S. cereale</i>	7.3	146.0	LT 522/6	11.7	129.3

all years, average FN values were higher in A-group of introgressive triticale lines (based on LT 176/10 triticale recipient parent) in relation to analogical B-group of lines developed with the participation of triticale LT 522/6. As could be seen from diagrams of dispersion (Fig. 1) among analyzed B-lines there were quite frequent lines with low FN (< 100) and to this group belongs as well some of lines distinguished by mature spikes sprouting tolerance (B20/1, B10/1-1) (Fig. 1). No one of the Tc/Tm lines showed FN values approximated in number to that of diploid wheat, however, 18 lines showed FN (means over 3 years) higher in relation to the best of triticale parental strain and to rye Dańkowskie Złote.

Table 2. Variation of the number of sprouted kernels per spike (SR) and Falling number (FN) in two series of introgressive lines different in genotype of triticale recipient parent (A - LT 176/10, B - LT 522/6).

Experiment Lines		SR mean	SD	CV [%]	FN mean	SD	CV [%]
I	A	3.80	3.098	81.53	108.43	43.019	39.67
	B	5.68	4.770	75.18	80.43	36.479	45.35
II	A	8.24	6.984	84.76	159.73	68.320	42.77
	B	9.55	7.336	76.82	99.58	42.749	42.93
III	A	15.43	15.392	99.75	170.12	66.850	39.30
	B	14.24	11.722	82.32	146.28	72.166	49.33

High FN (> 220) were characteristic for three of lines, all developed as introgressive progeny of LT176/10. Two of these lines (A9/2, A5/2) distinguished parallelly with tolerance of mature spikes to wetting but line A18/1 showed high sprouting in every mature spikes testing. This line is quite surprisingly found to have in every analysis high FN, even in the season with abundant rains (102 mm on July).

Generally, at any one of three vegetative seasons (different in climatic conditions, see Table 3) no one significant SR-FN correlation in A series of Tc/Tm lines existed. Also correlation coefficient based on averaged over 3-year values was very small and insignificant. In opposite to this, linear correlation coefficients estimated for SR-FN relationship showed good agreement between the FN and intact spikes SR for B series of Tc/Tm lines. Statistically significant coefficients of correlation were found in two of three vegetative seasons and average value over 3 years showed highly significant negative correlation (Table 3). From these results it is obvious that

the genes pool of triticale recipient parent showed strong influence on the FN in introgressive lines as well as on correlation of FN-SR. That is in opposite to the phenotypical expression of SR introgressed from *T. monococcum* which character showed to be independent on recipient parent genotype (Sodkiewicz and Tomczak 1994). It makes clear that in analyzed introgressive triticale lines the character of mature spikes SR was inherited in clearly different manner than kernels FN, showing different set of genes responsibility.

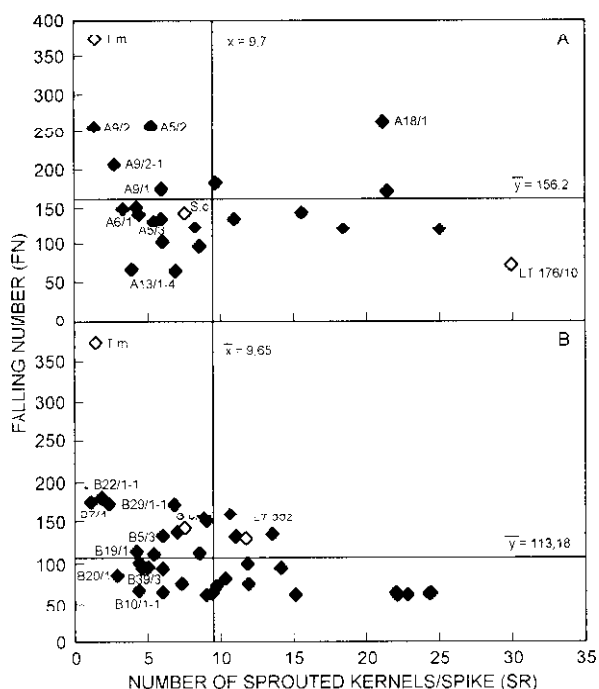


Fig. 1. A dispersion diagram for A series (A) and B series (B) of introgressive triticale lines classified according to the number of sprouted kernels in spike (SR) and kernels Falling Number (FN) averaged over experiment I, II, and III.

Table 3. Linear correlation coefficients for the number of sprouted kernels per spike (SR) and Falling Number (FN) estimated in 3-year experiment with two series of triticale introgressive lines differentiated in spike sprouting resistance (*, ** - statistically significant at $P < 0.05$ and $P < 0.01$, respectively).

Experiment	Rainfall in June	Lines	r (in experiments)	r (over 3 years)
I	54 mm	A	-0.048	-0.028
		B	-0.276	-0.509**
II	102 mm	A	-0.298	
		B	-0.392*	
III	73 mm	A	-0.193	
		B	-0.430*	

The level of starch decay evaluated by FN is still one of the commonly used method of determining the kernel damages caused by α -amylase activity. In opposite to the intact spikes sprouting character, in both series of introgressive triticale genotypes there were no found lines having FN comparable with that of diploid wheat. It means that genetic background of FN is more complicated in relation to SR inheritance.

Frequently seen independency of FN and SR (see lines B20/1, B10/1-1, A-lines generally) demonstrate the existence of other genetic systems significantly influencing the level of kernel starch damage. This observation paid attention on the phenomenon of α -amylase synthesis before spikes maturity in unripened and ungerminated kernels (Flintham and Gale 1988).

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