

## LATVIJAS SARKANĀ ĀBOLIŅA (*TRIFOLIUM PRATENSE* L.) ŠĶIRŅU ĢENĒTISKĀ PASPORTIZĀCIJA: VIETAS UN LAIKA IETEKME UZ ĢENĒTISKO DAUDZVEIDĪBU

**Bērziņa I., Zhuk A., Veinberga I., Rashal I. and Ruņģis D.**

Ģenētisko resursu centrā ir izstrādāta ģenētiskas pasportizācijas metodika Latvijas gēnu bankā (LGB) uzglabātām sarkanā āboliņa šķirnēm. Sarkanais āboliņš ir svešapputes augs un ņemot vērā, ka šķirnes ir audzētas un atjaunotas bez īpašām izolācijas metodēm, šķirnes robežās ir sagaidāma augsta ģenētiskā daudzveidība.

Izpētītas septiņas diploīdā sarkanā āboliņa šķirnes, kuras izveidotas trīs Latvijas selekcijas institūtos. Tika analizētas sēklas, kuras ievietotas LGB 1999.g. un 2000.g. Šķirnei 'Priekuļu 66' tika analizētas no VIR repatriētās sēklas (ievietotas VIR kolekcijā 1982.g.), Latvijas gēnu bankas sēklas (1999.g.), kā arī augi no Priekuļu lauka kolekcijas 2007. gadā.

Šāda dažādu avotu svešapputes augu ar augstu ģenētiskā daudzveidību analīze ļauj izpētīt ģeogrāfisko ietekmi uz ģenētisko daudzveidību (šķirnes izveidotas dažādos selekcijas institūtos), kā arī to izmaiņas laikā (repatriētās sēklas, LGB sēklas un lauku augi).

## GENETIC AND ENVIRONMENTAL EFFECT ON THE GRAIN QUALITY OF SPRING BARLEY

**Bleidere M.**

State Stende Cereal Breeding Institute, p. Dizstende, Talsi region, Latvia, LV-3258

phone: +371 26229659, e-mail: [maara.bleidere@stendeselekcija.lv](mailto:maara.bleidere@stendeselekcija.lv)

### Abstract

The grain quality of spring barley (*Hordeum vulgare* L.) determines the further opportunities of their utilization. This study investigated the effects of genotype and environment on the grain quality of spring barley. Fifty two spring barley genotypes were used for this investigation. Test weight, 1000 grain weight and grain composition (starch, crude protein, crude fat,  $\beta$ -glucans, crude fibre, crude ash) were evaluated. Field experiments were carried out at the State Stende cereal breeding institute from 2004 to 2006. The results were analyzed to synthesize the relative proportion of the influence various factors ( $\eta$  %) such as variety or year as well as the influence of climatic conditions (mean air temperature and the amount of rainfall) during the period of grain filling. The analysis of variance suggested that the fluidity of grain quality indices were more strongly ( $p < 0.01$ ) affected by their genotype. While measuring the significance of each single factor, the influence of genotype factor was found to be over 70% in starch, crude protein,  $\beta$ -glucans, crude fibre, 1000 grain weight and test weight. A significant environmental influence on grain quality was found for all characteristics except  $\beta$ -glucan and test weight. This was attributed to the varied meteorological conditions during the years of investigation in the the first part of grain filling period.

**Key words:** spring barley, grain quality, analysis of variance, meteorological conditions.

### Introduction

Barley is used for a wide range of uses. It is an important crop used for feed for different livestock species in many areas of the world and also in Latvia.

The most commonly used characteristics to describe the quality of barley in breeding are the physical traits and chemical composition of the grain. The major chemical components of barley grain are starch, protein and  $\beta$ -glucans. The other constituents of barley include ash and fiber components. Fiber components include pentosans, cellulose, and lignin. They are important to measure because of their contribution toward the total dietary fiber. Ash represents the mineral component of barley. The amount of energy available to an animal from grain depends on the relative proportion of each chemical constituent (Shewry and Morell, 2001). Variation in these proportions is genetically controlled (Eagles *et al.*, 1995), but it is also influenced by environmental

factors (O' Brien, 1999; Paynter and Young, 2004; Helm, 1992). Barley with plumper grains and a higher test weight should have a greater percentage of starch or energy in the grain and should be lower in fibre (Shewry and Morell, 2001).

The different components of a barley grain are synthesized in sequence. The growth of cariopsis after anthesis can be divided into two main stages, cell division and grain filling (Hay and Porter, 2006). The duration of grain growth depends strongly on temperature, therefore it is one of the most important factors affecting grain quality (Pasarella, 2005). Another important factor influencing the duration of growth is the stage of cereals and the quality of the water supply. Water deficit influences grain filling through the reduction in the assimilate supply (Paynter and Young, 2004; Conzalet *et al.*, 2007).

It was assumed that the difference between varieties grown at the same location were due to the genetics of the plant. Plant breeders also discovered that the environment plays a big role in the resulting quality of the grain. If the main effect on the quality factor is due to the environment then breeding would have very little influence.

The objective of this study was to determine the influence of genotype and environment on the grain quality traits for covered, hulless, two-row and six-row barley grown in different growing seasons. Emphasis was placed the interpretation of the role of meteorological variables in the period of grain filling on grain quality differences over the years of the study.

### Materials and Methods

52 barley genotypes were chosen which represented a broad range of germplasm (two-row, six-row, covered, and hulless) of different origin (Table 1). Thirty-eight genotypes of covered spring barley, from which 27 with two-row and 10 with six-row ear types, and 15 hulless genotypes were used in this study. Only two-row hulless genotypes were included in this investigation.

Table 1. Spring barley genotypes used in the study

Barley type	n	Genotype, origin country
Two-row, covered	27	Ansis, Abava, Sencis, Kristaps, Rasa, Linga, Idumeja, Balga, Ruja, Gate, Malva, Klinta (Latvia); Hanka, Annabell, Danuta, Justina, Polygena (Germany); Austrian early, Landsorte Aus Tirol (Austria); Primus II (Sweden); Lysimax (Denmark); Hatvani 45/25 (Hungary); Cork, Century (Great Britain); Lechtaler (Portugal); Grimmet (Australia); 379 (Chile)
Six-row, covered	10	Druvis (Latvia); Colsess IV, July (Denmark); B90A, RNB-367 (Nepal); Zoapila, Puebla (Mexico); IV/192 (Macedonia); Valluno (Bolivia); Chosen (North Korea)
Two-row, hulless	15	L 302 (Latvia); KM 2084 (the Czech Republic); SW 1291 (Sweden); McGwire, Gainer, Merlin, Candle (Canada); X-4 (Lithuania); 10250 (Russia); Orzo Nudo di Altamura (Italy); 2474, Clho 7799 (Guatemala); C.P.I. 22817 (Russia); Sumire Mochi (Japan), Wanubet (USA)

The genotypes were grown at the State Stende Cereal Breeding Institute from 2004 to 2006. The soil at the site was sod-podzolic sandy loam, humus content – 12-15 mg kg<sup>-1</sup>, soil pH<sub>KCl</sub> – 6.0-6.7, precrop – potatoes, available for plants P – 88-94 mg kg<sup>-1</sup> and K – 103-122 mg kg<sup>-1</sup>. The size of the plot was 2 m<sup>2</sup>, 2 replicates, and seed rate - 400 seeds per m<sup>2</sup>. The plots were fertilized with N60 P15 K40 kg ha<sup>-1</sup>.

Prior to the analysis, a representative grain sample from both replications was ground in a Perten cyclone mill to pass a 0.8 mm screen. Crude protein content (CP) (N x 6.25) was determined by the Kjeldahl method (LVS 277).  $\alpha$ -glucans content (BGL) was analyzed enzymatically following the barley grain procedures of the commercial kits from *Megazyme* (Megazyme International Ireland Ltd.). Starch content (STR) (ISO 10520), crude fiber content (FBR) (ISO 5498), crude fat content (FAT) (ISO 6492) and crude ash content (ASH) (LVS 276:2000) were determined. All chemical analyses were reported on a dry matter basis. The dry matter content of the ground grains was determined by oven-drying at 130 °C for 2 h. 1000 grain weight (TGW) (LV ST ZM43-95) and test weight (TW) (LVS ISO 7971-2) were analyzed.

The growing season of 2006 had a lower amount of precipitation and a higher daily temperature in comparison with 2004 and 2005. The climatic conditions for the first part of the growing season

(April and May) of 2006 can be characterized as having a heightened temperature and a reduced amount of precipitation. The mean air temperature in June of 2004 and 2005 was below the long-term data (1.7 and 0.6 °C respectively), but in 2006 it was above the long term data (1.1 °C). The highest amount of precipitation was observed in June of 2004 (199% of the long-term data). June and July of 2005 as well as June, July and August of 2006 were characterized by a reduced amount of precipitation. July of 2004 and 2006 was cool (0.8 and 0.4 °C under the long-term data). The meteorological conditions of the month August during the years of investigation were comparatively warm (from 0.5 to 1.9 °C above the long-term data).

The heading and maturity date were determined. The grain filling period from heading to maturity was calculated. As the starchy endosperm of the major storage tissue differentiates during the first part of grain filling the periods from heading to maturity were divided into two similar parts – 20 to 25 day periods depending on the length of this period in order to determine the difference of the mean daily air temperature and the sum of rainfall during the grain filling period during the years of the investigation. The amount of rainfall and the mean daily air temperatures for each period were calculated (Table 2).

Table 2. Meteorological differences during the grain filling period of spring barley, 2004-2006

Variable	2004	2005	2006
Average heading date	1.07	27.06	28.06
Average maturity date	14.08	16.08	6.08
Average length of grain filling, days	45	51	40
Mean daily air temperature, °C			
Grain filling period	16.4a <sup>1</sup>	17.0a	16.3a
1st part of grain filling	15.0a	17.8b	19.4c
2nd part of grain filling	17.6a	16.2b	13.1c
Sum of precipitation during grain filling, mm			
Grain filling period	117.6	168.4	50.4
1st part of grain filling	79.5	31.30	5.1
2nd part of grain filling	38.1	137.1	45.3

<sup>1</sup>means in each comparison between years followed by different letters are significantly different at the  $p \leq 0.05$  level.

Two-way ANOVA was performed using the program *Microsoft Excel*. According to the analysis of variance the proportion influence of relative factors ( $\eta$  %) measuring the relative importance of each single factor (genotype, year, residual or interaction) was calculated. Comparisons were carried out using the t-test to estimate differences between the years.

### Results and Discussion

According to the analysis of variance for the three year data, a significant difference ( $p$ -value  $\leq 0.01$ ) between genotypes in all investigated grain quality traits was observed when all genotypes were included in the analysis (Table 3). Differences were found, if the analysis of variance was performed based on type of barley: covered, hullless, two-row and six row. For covered and hullless barley the genotype as a factor was also significant at the high level of probability ( $p \leq 0.01$ ). For six-row barley the genotype influence for crude protein, starch,  $\beta$ -glucans, crude fibre, 1000 grain weight and test weight was statistically significant ( $p \leq 0.01$ ), but for crude fat and crude ash content it was of no significance (Table 3). It means that there was no significant differences between six-row genotypes in both mentioned parameters.

Table 3. Analysis of variance for grain quality traits of different types of spring barley<sup>1</sup>

Source	CP	STR	FAT	BGL	ASH	FBR	TGW	TW
Proportion of factors influence ( $\eta\%$ ) and probability of F								
<i>Genotype</i>								
All genotypes	78.5** <sup>2</sup>	72.8**	67.0**	84.0**	52.1**	90.7**	80.6**	95.5**
Covered	63.2**	68.9**	57.4**	59.3**	33.4**	52.5**	76.9**	87.4**
Hulless	74.4**	73.1**	73.8**	79.5**	71.2**	67.1**	86.6**	84.7**
Two-row	49.5**	62.3**	62.6**	47.2*	23.2**	28.9	60.3**	80.9**
Six-row	75.7**	51.4* <sup>3</sup>	32.3	69.4**	41.4	67.0**	80.7**	65.1**
<i>Year</i>								
All genotypes	12.1**	6.4**	3.7**	0.4	24.6**	5.5**	8.9**	0.2
Covered	13.1**	3.2*	7.6**	3.3	39.4**	32.0**	9.8**	1.4*
Hulless	15.9**	6.6*	3.2	0.1	10.1**	17.2**	7.7**	2.2
Two-row	18.1**	13.5**	7.8**	3.2	59.0**	19.5**	19.5**	2.4*
Six-row	11.1**	14.1*	14.3	5.2	10.5	17.1**	5.9**	3.2
<i>Interraction</i>								
All genotypes	9.4	20.6	24.1	15.5	23.1	3.7	10.4	4.2
Covered	23.6	27.7	35.0	37.3	27.1	15.3	13.1	11.1
Hulless	9.6	20.1	22.9	20.3	18.6	15.2	5.6	13.0
Two-row	32.3	24.2	29.6	49.5	17.7	51.4	20.1	16.6
Six-row	13.1	34.4	53.3	25.2	48.1	15.7	12.3	31.5

<sup>1</sup>CP-crude protein, STR-starch, FAT-crude fat, BGL- $\beta$ -glucan, ASH-crude ash, FBR-crude fibre, TGW-1000 grain weight, TW-test weight; <sup>2</sup>  $p \leq 0.01$ ; <sup>3</sup>  $p \leq 0.05$ ;

This parameter showed that starch (72,8 %), crude protein (78,5 %),  $\beta$ -glucans (84 %), crude fibre (90,7 %), 1000 grain weight (80,6 %) and test weight (95,5 %) were governed mainly by the genotypes (Table 3). The results showed that genotype as a factor had a lower influence on the grain quality traits of different types of barley. Only for hulless barley it was higher in starch, crude fat, crude ash and 1000 grain weight. This indicated the highest genotypic variability among these traits for this specific type of barley.

The influence of a specific year was significant ( $p \leq 0.01$ ) for all grain quality traits except  $\beta$ -glucans and test weight when all genotypes were included in the analysis (Table 3). The influence of a year or growing season as a factor in the variance of grain quality traits was relatively small in comparison to the genotypic effect. The influence of density factors varied from 0.2% for the test weight to 12.1% for crude protein when all genotypes were included in the analysis. The relative value of a year as a factor in the final statistical dispersion increased when the quality traits were analyzed depending on the type of barley. The differences between growing seasons were not significant for hulless barley in crude fat,  $\beta$ -glucans and test weigh. For six-row barley it was not significant for crude fat,  $\beta$ -glucans, crude ash and test weight, but for two-row barley only for  $\beta$ -glucans.

The analysis of variances also revealed the interactions of genotype and environment for all measured characteristics (Table 4).

Table 4. Average values of grain quality traits of spring barley (2004-2006)

Grain quality indice	2004	2005	2006
Crude protein, g kg <sup>-1</sup>	137.3a <sup>1</sup>	132.2b	150.8c
Starch, g kg <sup>-1</sup>	619.4a	624.9a	606.8b
Crude fat, g kg <sup>-1</sup>	23.0a	24.6b	23.7a
$\beta$ -glucans, g kg <sup>-1</sup>	42.7a	42.1a	43.2a
Crude ash, g kg <sup>-1</sup>	24.3a	21.6b	22.2c
Crude fibre, g kg <sup>-1</sup>	42.3a	43.8b	36.9c
1000 grain weight, g	45.5a	41.8b	44.4c
Test weight, g l <sup>-1</sup>	701.8a	695.6a	697.0a

<sup>1</sup> means in each comparison between years followed by different letters are significantly different at the  $p \leq 0.05$  level.

The amount of interactions between the genotype and the year was relatively small for crude protein (9.4%), crude fibre (3.7%), 1000 grain weight (10.4%) and test weight (4.2%) if the analysis of variance was performed for all genotypes. For other traits it was from 15.5 to 24.1% of the total variance. The significant influence of the density factors was for two-row and six-row barley for all evaluated quality traits. The results showed that crude ash was a trait where genotype, year and interaction determined the variation of this trait at the same rate. This means that this trait depended more on the genotype reaction to different environments.

As the t-test indicated, the mean comparisons for grain crude protein, crude ash, crude fibre and 1000 grain weight was significant for all the years of the investigation (Table 4). Barley grown in 2004 had significantly higher crude ash, 1000 grain weight and test weight than barley grown in 2005 and 2006. Barley had the highest starch content and crude fibre content in the grain in 2005. In this year the barley genotypes formed significantly lower 1000 grain weight. During the growing season of 2006 barley had a significantly lower starch content and crude fibre content, however the barley grain had the highest crude protein content in comparison to other years. Grain test weight and  $\beta$ -glucans did not show significant differences through the test years.

This study was undertaken to determine whether the difference in grain quality measurements was due to the effects of the genotype (the variety) or the environment (meteorological variables) as well as the interaction between of these both factors. The genotypic factor was found to be the primary one that significantly effected grain quality. Differences in grain quality especially concerning starch and protein was also related to genetic differences in how each cultivar accumulated these components during grain development (Savin and Molina-Cano, 2002). The significant effect of genotypes on the variability of grain quality parameters could be primarily due to the difference between types of barley - covered, hulless, two-row and six-row. According to the earlier results (Bleidere and Grunte, 2007), when grain quality was compared between different types of barley, the grain of hulless barley had significantly higher mean values of crude protein, crude fat and significantly lower crude fibre and crude ash values than the covered ones. Six-row barley contained a significantly higher amount of crude protein, crude fibre, crude ash, but less starch than the two-row barley. Also Bach Knudsen *et al* (1987) examined the chemical composition of a range of barley grown during three years in Denmark. The analysis of variance identified highly significant differences between varieties concerning protein, starch and the total dietary fibre content.

The variation of grain quality of the same variety grown in different years is caused mainly by the difference in environmental conditions. In this study phenological and meteorological data were used to analyse the differences in grain quality parameters between the different years of the investigation. The actual process of grain formation and filling take place between the heading and maturity stages (Hay and Porter, 2006). The average heading and maturity dates for spring barley showed differences over the period examined (Table 1). The longest grain filling period in 2005 was associated with high amounts of rainfall during this period. In 2005 and 2006, the first part of the grain filling period took place under exceptionally hot conditions. Additionally, in 2006 this period was notably dry. M. Wallwork *et al.* (1998) indicated that if a short period of high temperature occurs at a certain point in the grain filling period, it may affect one or more components that are being synthesized concurrently and result in a different composition of the mature grain. The high temperatures and the stress of moisture can limit the amount of grain fill operating through the metabolism of starch in the grain. It is because the accumulation of starch is more sensitive to high temperatures than to the accumulation of nitrogen, which frequently determines increases in the grain nitrogen proportion and thus result in higher protein contents (Schelling *et al.*, 2003). The hot and dry period during the year of 2006 corresponded to the period of cell division in starchy endosperm. Such conditions shortened the length of this period thus influencing the accumulation of starch. Due to this reason the season of 2006 resulted in the lowest starch and the highest protein content (Table 1). These factors translated to the slight reduction of the feeding value of barley, as feed grains are used mainly as the source of energy.

The most likely factor contributing to the interaction of the effects on grain quality traits, especially for the grain weight and the test weight were attributed to the dissimilar resistance of genotypes to lodging (caused by increased plant height and excessive rain at the time of grain filling). During the second part of grain filling in 2005 (from July 25 to August 16) there were

particularly high amounts of precipitation that caused early lodging for part of the genotypes and consequently decreased the mean grain weight and test weight. This could be also the main reason for the significantly higher crude fibre content during this year compared to the other years of investigation.

### Conclusions

The analysis of variance suggested that all grain quality traits were more strongly ( $p < 0.01$ ) influenced by the genotype. The proportion of influencing factors ( $\eta$  %) showed starch, crude protein,  $\beta$ -glucans, crude fibre, 1000 grain weight and test weight to be governed mainly by the genotype. The relative magnitude of the genotype as a factor increased when the quality traits analyzed depended on the type of barley. The significant environmental influences for all characteristics except for  $\beta$ -glucans and test weight were attributed to the different meteorological conditions between the different years of investigation during the grain filling period. The factor that was most likely contributing to the interaction of grain quality traits, especially for grain weight and test weight, was attributed to the diverse resistance of genotypes to lodging. The use of genetic variation in endosperm composition and the relative contribution of genotype and environment illustrate the opportunities that exist to improve nutritional value by direct breeding efforts.

### References

1. Bach Knudsen K., Eggum E., Jakobsen B.O. (1987) Nutritive value of Danish-grown barley varieties II. Effect of carbonhydrates composition on digestibility of energy and protein, *Journal Cereal Science*, 6, 187-195.
2. Bleidere M., Grunte I. (2007) Grain chemical composition of spring barley genotypes, International scientific conference proceedings „Research for rural development 2007”, Jelgava, May 16-17, 36-41.
3. Conzales, A., Martin, I., Ayerbe, L. (2007) Response of barley genotypes to terminal soil moisture stress: phenology, growth and yield. *Australian Journal of Agricultural Research*, 58, 29-37.
4. Eagles H.A., Bedggod A.G., Panozzo J.F., Martin P.J. (1995) Cultivar and environmental effects on malting quality in barley, *Australian Journal of Agricultural Research*, 46, 831-844.
5. Hay R.K.M., Porter J.R. (2006) Physiology of crop quality, In: *Physiology of crop yield*. Hay R. And Porter J.R.(ed.) Blackwell Publishing, Oxford, -328.
6. Helm J.H. (1992) Cultivar and growing location effects on composition of barley grains, Barley for food and malt, Proceedings of International simposium, September 7-10, Sweden, Uppsala, 308-312.
7. O' Brien L. (1999) Genotype and environment effects on feed grain quality, *Australian Journal Agricultural Research*, 50, 703-719.
8. Passarella V., Savin R., Slafer G. (2005) Breeding effects on sensitivity of barley grain weight and quality to events of high temperature during grain filling, *Euphytica*, 141, 41-48.
9. Paynter B. H., Young K. J. (2004) Grain and malting quality in two-row spring barley are influenced by grain filling moisture, *Australian Journal Agricultural Research*, 55, 539-550.
10. Schelling K., Born K., Weissteiner C., Kunbauch W. (2003) Relationships between yield and quality parameters of barley (*Hordeum vulgare L.*) and phenological and meteorological data, *Journal of Agronomy and Crop Science*, 189, 113-122.
11. Shewry P.R., Morell M. (2001) Manipulating cereal endosperm structure, development and composition to improve end-use properties, *Advanced Botany Research*, 34, 165-234.
12. Swantson J.S., Ellis R.P., Perez-Vedrell A., Voltas J., Molina-Cano J.L. (1997) Paterns of barley grain development in Spain and Scotland and their implication for malting quality, *Cereal Chemistry*, 74, 456-461.
13. Wallwork M.A.B., Logue S.J., MacLeod L.C., Henner C.F. (1998) Effect of the period of high temperature during grain filling on starch synthesis in developing barley grain, *Australian Journal of Plant Physiology*, 25, 173-181.
14. Savin R., Molina-Cano J.L. (2002) Changes in malting quality and its determinants in response to abiotic stress, In: Barley Science. Recent advances from molecular biology to agronomy of yield and quality, Slafer A.G.(ed.), The Haworth Press, Inc., USA, 523-550.

## GENOTIPA UN VIDES IETEKME UZ VASARAS MIEŽU GRAUDU KVALITĀTI

**Bleidere M.**

Vasaras miežu (*Hordeum vulgare* L.) graudu kvalitāte būtiski nosaka to tālākās izmantošanas iespējas pārstrādē. Pētījuma mērķis bija noteikt genotipa un vides ietekmi uz miežu graudu kvalitāti. Pētījums veikts Valsts Stendes graudaugu selekcijas institūtā no 2004. līdz 2006. gadam. Piecdesmit diviem genotipiem novērtēja tilpummasu, 1000 graudu masu un graudu ķīmisko sastāvu (cieti, kopproteīnu, koptaustus,  $\beta$ -glikānus, kokšķiedru un koppelus). Analizēja faktoru (šķirne, gads) ietekmes īpatsvaru ( $\eta\%$ ), kā arī meteoroloģiskos apstākļus (vidējā gaisa temperatūra un nokrišņu summa) graudu veidošanās periodā. Dispersijas analīze liecina, ka visu novērtēto graudu kvalitātes rādītāju mainību būtiski ( $p \leq 0.01$ ) ietekmē genotips. Genotipa kā faktora ietekmes īpatsvars ( $\eta\%$ ) virs 70% bija cietei, kopproteīnam,  $\beta$ -glikāniem, kokšķiedrai, 1000 graudu masai un tilpummasai. Konstatēta būtiska gada ietekme uz graudu kvalitātes mainību visām pazīmēm, izņemot  $\beta$ -glikāniem un tilpummasai. To nosaka starp gadiem atšķirīgie meteoroloģiskie apstākļi graudu veidošanās periodā.

## THE EFFECT OF TILLER NUMBERS IN WHEAT HAPLOID PRODUCTION

**Brazauskas G.**

Lithuanian Institute of Agriculture, LT-58344 Akademija, Kėdainiai r., Lithuania

phone: +37034737149, e-mail: [gintaras@lzi.lt](mailto:gintaras@lzi.lt)

### Abstract

Winter wheat doubled haploid production is widely used not only for plant breeding acceleration but also in basic research, such as genetic mapping, haploid transformation and artificial seed production. It is essential to identify all factors which determine haploid system productivity. We have studied the effect of tiller numbers on embryo development and haploid regeneration in wheat x maize haploid production system. Four winter wheat F<sub>1</sub> hybrid lines were pollinated with the maize variety 'Golden Bantam' pollen. Embryo formation and haploid regeneration data were collected from the first five tillers in each plant. The data showed a clear tendency of reduction in embryo formation frequency on the successive tillers. There were 17.5% and only 12.4% embryos on average formed on the main and fourth tillers, respectively. However, two-way ANOVA analysis showed these differences to be insignificant ( $p > 0.05$ ) and only the genotype had a significant ( $p < 0.01$ ) effect on embryo formation. There was no clear effect by both genotype and tiller numbers on haploid regeneration frequency. The overall embryo formation and haploid regeneration frequency in this experiment was 14.6% (504 embryos) and 68.1% (367 haploids), respectively. The results obtained in this study indicate that as many as five tillers per plant can be used in wheat x maize crossing systems with no significant tiller effect on the efficiency of the haploid production system.

**Key words:** wheat x maize, haploid, tiller number.

### Introduction

Winter wheat doubled haploid production is widely used not only for plant breeding acceleration but also in basic research, such as genetic mapping, haploid transformation and artificial seed production. Several techniques exist for wheat haploid development. These include crossing with maize (Laurie and Bennet, 1988), anther culture (Last and Brettell, 1990) and isolated microspore culture (Gustafson *et al.*, 1995). Wheat x maize crossing is the system of choice in winter wheat breeding programs due to lower genotype dependency and albinism avoidance (Snape, 1998). However, wheat x maize crossing is a time consuming and costly technique. It requires manual application of maize pollen and subsequent embryo excision. Ways to determine the factors underlying the efficacy of the method are being searched for.

Various factors are related to the differences in wheat haploid formation after crossing with maize. Campbell *et al.*, (1998) determined that both temperature and light intensity influence embryo formation in the wheat x maize system. This effect was later associated with the abnormalities in