Euphytica
DOI 10.1007/s10681-013-1052-6

Effect of genotypic, meteorological and agronomic factors on the gluten index of winter durum wheat

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Received: 23 July 2013 / Accepted: 13 December 2013

Abstract The determination of the gluten index is a widely used method for analysing the gluten strength of bread wheat and spring durum wheat genotypes. The present work was carried out to study the effect of the genotype, meteorological factors (temperature, precipitation and number of days with $T_{max} \geq 30$ °C) and agronomic treatments (N fertilisation and plant protection) on the gluten index of winter durum wheat varieties and breeding lines. The results indicated that the gluten index had little dependence on the environment, being determined to the greatest extent by the genotype. Compared with varieties having weak gluten, those with a strong gluten matrix responded less sensitively to changes in environmental conditions. Among the meteorological factors, high temperature at the end of the grain-filling period caused the greatest reduction in the mean gluten index of three varieties ($R^2 = 0.462$), while the fertiliser was found to be a significant factor affecting the gluten strength of winter durum wheat varieties. Using selection based on the gluten index, the gluten strength of winter durum wheat lines can be improved sufficiently to make them competitive with high quality spring varieties.

Keywords *Triticum turgidum* ssp. *durum* · Gluten strength · Breeding for quality · Pasta-making quality

Electronic supplementary material The online version of this article (doi:10.1007/s10681-013-1052-6) contains supplementary material, which is available to authorized users.

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Introduction

Durum wheat has a number of traits which make it ideal for pasta-making. Due to its high yellow pigment content, attractive products can be manufactured without the addition of eggs (Matsuo and Irvine 1967). It also has a high protein content, while its strong gluten matrix retains the starch molecules during cooking (Feillet 1984), with the result that the surface of the pasta does not become sticky and the pasta keeps its shape (Dexter and Matsuo 1980).

The cooking properties and sensory value of pasta are jointly determined by a number of factors. The choice of raw material is of decisive importance. Both the quantity and quality of the protein and the nature of the starch exert an effect on the properties of the end-product (Dexter and Matsuo 1977, 1980; Malcolmson et al. 1993). In addition to suitable raw materials, the processing and cooking technology also influence the palatability and digestibility of cooked pasta (Petitot et al. 2009; Bruneel et al. 2010). The results of electron microscope studies (Cunin et al. 1995) showed that interactions between the gluten matrix and the starch

Published online: 21 December 2013

determine the quantity of starch molecules reaching the pasta surface during the cooking process, and thus the stickiness of the pasta.

The gluten structure, including the strength of the matrix, is thus an important component of pasta quality. Gluten strength can be determined using several techniques. The laboratory equipment widely used for rheological measurements on bread wheat can also be used to analyse durum wheat samples (Sissons et al. 2012). In addition to measurements performed with the farinograph, mixograph and alveograph, various other methods are also suitable for the determination of gluten strength, including visco-elastograph analysis, the SDS sedimentation test and manual tests of elasticity (D'Edigio et al. 1990). The determination of the gluten index (GI), based on the methodology elaborated by Perten (1990), is also a useful technique for use in durum wheat breeding programmes. A modified version of this method was first used for the analysis of durum wheat wholemeal and semolina samples by Cubbada et al. (1992), who found that the GI was closely correlated with the results of manual gluten quality determinations and was in close correlation with the SDS sedimentation volume ($r = 0.78$–0.81) and with the alveograph W value. It is possible to perform the measurements even on wholemeal, and only 20 g samples are required for the analysis, so the method is particularly suitable in breeding programmes for analysing samples from early generations. Compared with the SDS sedimentation test, the results are less dependent on the protein concentration, enabling breeders to carry out objective selection (Clarke et al. 2010). Correlations between the GI and other rheological traits have been reported by a number of authors. Edwards et al. (2007) demonstrated close correlations with the alveograph W value, the unextractable polymeric protein fraction and the mixograph mixing time, and Lerner et al. (2004) with the energy level and tolerance determined using a farinograph.

The GI is a stable technological quality trait, dependent on the genotype (Ames et al. 1999). Values of the GI recorded using semolina were not significantly influenced by nitrogen fertilisation (Ames et al. 2003). The GI is a highly heritable genetic trait. In experiments performed by Clarke et al. (2000) the heritability of 120 progenies from three crossing combinations ranged from 0.84 to 0.93. These results were confirmed when the tests were repeated with a larger number of genotypes (six combinations, 398 lines; $h^2 = 0.84$–0.95) (Clarke et al. 2009).

The first determination of GI in durum wheat was reported 20 years ago, yet very few data have been published during the intervening period. This is particularly true of winter durum varieties. Winter durum wheat represents a special group of genotypes within the durum wheat species, and its cultivation is restricted primarily to countries in Eastern and Central Europe (Palamarchuk 2005). The first autumn-sown varieties appeared in Hungary in the 1980s, but these were still facultative types, which yielded 30–40 % more when sown in autumn than in spring (Beke and Barabás 1981). Testing of the first genuine winter durum wheat lines began in Martonvásár in 1982. These originated from the Odessa breeding programme and had excellent cold tolerance, but their technological quality was poorer than that of spring durum wheat varieties (Szunics et al. 1998). Improving cold tolerance and technological quality simultaneously is no easy matter. The varieties used as sources of cold tolerance did not have satisfactory gluten strength, so high quality facultative durum wheat varieties were also included in the crossing programme. The present paper reports on the results achieved in improving the gluten strength of winter durum wheat, and also discusses the effect of genotype, meteorological factors and agronomic treatment combinations on the GI.

Materials and methods

The work included three groups of experiments with the following aims: (1) To assess variability for GI in the winter and facultative durum wheat gene pool. (2) To identify meteorological factors influencing the GI. (3) To investigate the effect of agronomic treatment combinations on the GI of durum wheat varieties. All the experiments were set up on the same field in Martonvásár (47°18′N/18°49′E). According to the laboratory analysis of samples taken from the ploughed layer (1–20 cm) of the chernozem soil with forest residues, the soil of the selected area was sufficiently homogeneous. The topsoil, which contained no lime or damaging salts, had a weakly acidic pH and a loamy texture. Based on the humus content, it had moderate N supplies, while the AL-soluble values showed P contents close to the borderline between the

Table 1 Meteorological parameters and length of the 2000–2012 durum wheat vegetation periods in Martonvásár

Vegetation period	Precipitation (mm)	Mean temperature (°C)	No. of hot days[a]	Day of the year at		Main meteorological stress factor(s)
				Sowing	Harvest	
1999/2000	355.1	8.67	27	284	178	Hot and dry grain-filling period
2000/2001	397.5	8.79	5	271	188	Dry grain-filling period
2001/2002	244.0	8.82	18	272	179	Cold in December, terminal heat stress
2002/2003	200.4	7.44	29	283	178	Cold winter, hot and dry grain-filling period
2003/2004	416.8	7.42	3	281	201	Cold in January
2004/2005	442.6	7.11	10	281	196	Rainy harvest
2005/2006	378.5	7.13	17	283	191	Cold in January, terminal heat stress
2006/2007	147.9	10.32	25	285	176	Drought, terminal heat stress
2007/2008	430.8	8.33	19	284	184	Excessive amount of precipitation
2008/2009	283.6	8.20	12	284	182	Dry grain filling period
2009/2010	555.0	7.56	11	281	195	Excessive amount of precipitation
2010/2011	228.3	7.68	7	287	188	Cold in December, terminal heat stress
2011/2012	202.5	8.13	22	283	180	Drought, terminal heat stress

[a] $T_{max} \geq 30$ °C

Table 2 Summary of experiments aimed at studying the effect of genotype, meteorological factors and agronomy on the gluten index of winter durum wheat varieties and breeding lines

Experiments	Number of			Plot size
	Genotypes	Years	Replications	m^2
1. Assessment of variability for gluten index	70	3	2	2
2. Meteorological factors influencing the gluten index	3	13	2	6
3. Effect of agronomic treatments on gluten index	3	3	3	10

good and very good categories (200 mg kg^{-1}) and good K supplies. With regard to microelements, the soil had a low Zn content, but good supplies of Cu.

Sowing was performed with HEGE-80 or HEGE-90 seed drills (Hans-Ulrich Hege GmbH und Co., Waldenburg, Germany) and in all the experiments the plant density was that recommended in Hungary (500 seed/m^2). The crop was protected against weeds and insect pests throughout the growing season, but no fungicide was applied, except in the experiment set up for testing the fungicide effect as well as other agronomical factors. The plots were harvested at full maturity with a plot combine (Wintersteiger AG, Ried, Austria).

The data are based on GI measurements carried out on durum wheat samples harvested between 2000 and 2012. The meteorological parameters of the vegetation periods and the most important meteorological stress factors are presented in Table 1.

Each of the four experiments (Table 2) involved different genotypes and field methodology, so the material used for the quality analyses will be discussed separately.

The GI of each winter durum wheat sample was determined from semolina on the basis of the ICC158 standard (ICC 1995) using a Perten Glutomatic 2200 instrument and a Perten 2015 Centrifuge (Perten Instruments AB, Hägersten, Sweden). The semolina required for the analysis was prepared using a Brabender Junior laboratory mill (Brabender GmbH & Co. KG, Duisburg, Germany), converted as suggested by Vasiljevic et al. (1977). The removal of the bran and separation according to particle size were carried out on a Retsch sieve series (Retsch GmbH, Haan, Germany). The 160–315 µm fraction was further cleaned with a Chopin Semolina Purifier (Chopin Technologies, Villeneuve-la-Garenne, France). From 2010 onwards the durum wheat semolina samples were prepared using a Chopin CD2 laboratory mill, but the semolina was cleaned using the instrument described above, so the particle size of samples used before and after 2010 was the same.

Assessment of variability for GI in the winter and facultative durum wheat gene pool

A total of 70 varieties and breeding lines from the durum wheat breeding programmes of ten countries (Hungary, Germany, Austria, Ukraine, Romania, Croatia, Italy, Slovakia, Russia and Turkey) were tested in the experiments. The number of varieties from each country ranged from 1 (Russia) to 16 (Hungary). The experiments were set up in two replications in three consecutive years with contrasting weather conditions (2010, 2011 and 2012) with the aim of screening the available winter and facultative durum wheat gene pool and identifying possible new sources for quality improvement. The varieties making up the collection were thus sown in small plots measuring 2.0 × 1.0 m, with a row distance of 20 cm. The data were analysed using the Mixed Model (REML) module of GenStat 15th Edition (VSN International Ltd, Hemel Hempstead, UK) as described by Miedaner and Longin (2013).

Identification of meteorological factors influencing the GI

The GI data recorded between 2000 and 2012 for three winter durum wheat varieties with different gluten contents and gluten strength were used for the calculations. The variety 'Parus', which originated from Odessa, Ukraine and has weak gluten, has been used as a check variety ever since durum wheat experiments were begun in Martonvásár. The Martonvásár-bred winter durum wheat variety 'Mv Makaróni', registered in 2001, also has poor gluten strength. 'Mv Pennedur', however, also bred in Martonvásár and registered in 2011, is a winter durum wheat variety with strong gluten. The determination of the GI was carried out in two replications on samples from a field variety trial where the plot size was $6 \times 1.05\ m^2$, with a row distance of 15 cm.

The meteorological data used for the analysis were obtained from the database of the automatic measuring station set up in Martonvásár, which is also used by the Hungarian Meteorological Service. Tests were made on the effect of rainfall and temperature (monthly totals and means from November to March and 10-day totals and means from April to July 10th) and of the number of heat days ($T_{max} \geq 30$ °C; totalled until July 10th).

The data were analysed using the Mixed Model Analysis module of SPSS 16.0 software (SPSS Inc., Chicago, IL, USA). Year and Genotype were treated as fixed factors and the Block as a random factor. Correlations between the meteorological data and the GI of the varieties were determined on the basis of Pearson's correlation coefficient and stepwise regression using SPSS 16.0. Mean values of the two replications were used for the correlation and regression analysis.

Effect of agronomic treatment combinations on the GI of durum wheat varieties

The three-factorial small-plot field experiment was set up in a split–split-plot design with three replications in October 2009, 2010 and 2011. The whole plots contained six different levels of N supplies [0 kg N ha^{-1}; 50, 100 and 150 kg N ha^{-1} applied in a single dose in early spring; 70 + 30 and 120 + 30 kg N ha^{-1} applied in two doses, in early spring (GS23-25) and at the second node stage (GS32)]. Within each whole plot, five spraying treatments were applied in a random manner as subplots (1. untreated control; 2. treatment of spikes only, using a sprayer with single-jet nozzles; 3. treatment of both leaves and spikes, using a sprayer with single-jet nozzles; 4. treatment of spikes only, using a sprayer with twin-jet nozzles; 5. treatment of both leaves and spikes, using a sprayer with twin-jet nozzles). In treatments 3 and 5 the leaves were sprayed with a 1 L ha^{-1} dose of Amistar Xtra (200 g L^{-1} azoxystrobin + 80 g L^{-1} cyproconazole) in the first node stage (GS31), while in treatments 2–5 the spikes were sprayed with 2 L ha^{-1} Cherokee (50 g L^{-1} cyproconazole + 62.5 g L^{-1} propiconazole + 375 g L^{-1} chlorothalonil) fungicide at heading (GS59-61). The third factor (sub-subplots) was represented by three winter durum wheat varieties with different levels of gluten strength (1. 'Mv Makaróni': weak gluten; 2. 'Mv Hundur': intermediate; 3. 'Mv Pennedur': strong gluten). The data from each year and from the three years combined were analysed using the Mixed Model Analysis module of SPSS 16.0 software (SPSS Inc., Chicago, IL, USA) as described by Virk et al. (2009). The REML mixed model had two parts:

(i) Fixed model = Constant + Fertiliser level (N) + Fungicide treatment (F) + Genotype (G) + N × F + N × G + F × G + N × F × G
(ii) Random model (single year) = Replication, or
(iii) Random model (3 years combined) = Year + Year × Replication

Results

Assessment of variability for GI in the winter and facultative durum wheat gene pool

Measurements on the GI of varieties and breeding lines originating from the winter and facultative durum wheat breeding programmes of ten countries were made in 3 years with very diverse rainfall supplies (558 mm precipitation during the vegetation period in 2010, 228 mm in 2011 and 203 mm in 2012). The mean, maximum and minimum values and the corresponding variance components for the 70 winter durum wheat varieties and breeding lines tested between 2010 and 2012 are presented in Table 3, for the individual years and overall. The GI values for the breeding lines and for the Hungarian and foreign varieties ranged widely between 1.51 and 96.37 (Fig. 1). Despite the substantial differences in weather conditions the range had a similar order of magnitude each year. Genotypic variances were significantly ($p < 0.001$) different from zero. Variances due to the genotype × environment interaction and residual error were much lower than genotypic variances in each year and in the 3 years combined, resulting in high repeatability values (0.928–0.998).

The GI has been a selection criterion in the Martonvásár durum wheat breeding programme since 2000. The gluten strength of varieties tested in state variety trials prior to 2000 was very variable. On the basis of the GI, Martondur 1 and Mv Maxidur were classified in the good and excellent categories in most years, but the gluten quality of Martondur 2 and Mv Makaróni was poorer than average. The first durum wheat varieties for which the GI was a selection criterion in all stages of the breeding process were released in 2011. Based on the classification system of Cubbada et al. (1992) the gluten quality of Mv Hundur was average or above average, while that of Mv

Table 3 Descriptive statistics of the gluten index, wet gluten content, yellow index, heading time, plant height and leaf spot resistance, and the variance components (σ^2_G = genotypic; σ^2_{GY} = genotype × year interaction; σ^2_e = error variances) of 70 winter durum wheat genotypes. Martonvásár, 2010–2012

	Gluten index	Wet gluten content (%)	Yellow index[a]	Heading time[b]	Plant height (cm)	Leaf spot score[c]
Mean	52.675	43.305	20.908	140.100	91.250	3.507
Minimum	1.509	33.350	14.970	131.000	68.000	0.000
Maximum	96.366	63.350	28.220	150.000	124.000	8.000
Standard deviation	27.485	5.056	2.810	3.744	11.497	2.321
σ^2_G	590.304***	12.511***	6.979***	2.413***	43.491***	2.204***
σ^2_{GY}	28.097	6.391	0.177	0.310	27.547	0.944
σ^2_e	76.867	4.103	0.611	1.507	0.047	1.191
Repeatability	0.944	0.782	0.964	0.799	0.759	0.674
r_P^d		−0.484**	0.482**	0.047	−0.139*	0.139*

*, **, *** $p < 0.05$, 0.01 and 0.001, respectively

[a] Minolta b* value measured using a Minolta CR-300 Chromameter from semolina

[b] Day of the year

[c] 0–9 scale, 0 = free of symptoms, 9 = very sensitive

[d] Phenotypic correlation coefficients estimated between the best linear unbiased estimators (BLUEs) of gluten index and other traits

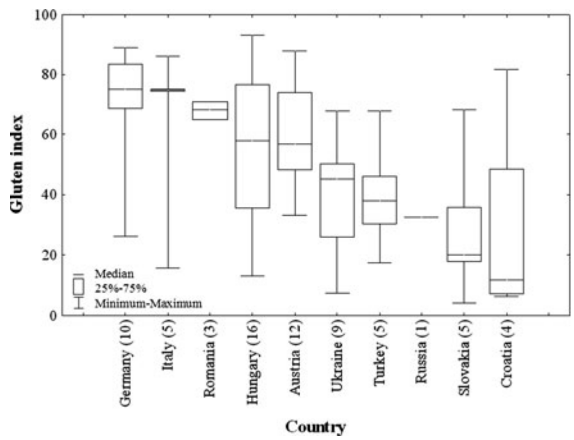

Fig. 1 Gluten index of winter and facultative durum wheat varieties and breeding lines from various countries (number of lines tested in *parenthesis*). Martonvásár, 2010–2012

Pennedur was excellent in 2010 and 2011 and very good in 2012.

Table 4 Wet gluten content (WG %) and gluten index (GI) of winter durum wheat varieties. Martonvásár, 2000–2012

Year	Parus		Mv Makaróni		Mv Pennedur	
	WG %	GI	WG %	GI	WG %	GI
2000	28.85	2.77	36.25	4.95	40.10	85.25
2001	41.15	50.39	38.00	38.13	39.65	93.08
2002	37.60	0.80	41.05	4.48	42.15	69.87
2003	45.65	11.06	40.85	22.14	39.05	89.87
2004	36.60	32.13	40.30	31.12	37.50	96.63
2005	34.25	1.16	36.55	1.89	38.50	91.44
2006	33.75	7.33	34.60	29.78	39.25	79.73
2007	31.55	10.87	36.00	22.82	36.80	85.86
2008	29.75	49.31	33.30	10.78	34.05	88.13
2009	21.00	59.49	24.60	68.54	25.00	98.61
2010	35.20	3.41	36.60	15.95	30.85	77.63
2011	38.15	6.14	39.20	21.43	37.25	94.36
2012	32.25	8.92	32.80	15.86	33.35	80.39
Mean	34.29	18.75	36.16	15.86	36.42	86.99
Range	24.65	58.69	16.45	66.65	17.15	28.27

Identification of meteorological factors influencing the GI

The GI of three winter durum wheat varieties ('Parus', 'Mv Makaróni' and 'Mv Pennedur') was recorded in 13 consecutive years (2000–2012). The results of REML analysis clearly showed the primary influence of the variety ($F_G = 473.43$***). Nevertheless, the year effect ($F_Y = 14.29$***) and the year × genotype interaction ($F_{Y \times G} = 3.19$***) were also significant, suggesting the importance of identifying the meteorological factors that influence this trait. The annual wet gluten content and GI data for the varieties are presented in Table 4. The correlation coefficient between wet gluten content and GI was not significant in the case of 'Parus' ($r = -0.326$), 'Mv Pennedur' ($r = -0.295$) and the mean of the three varieties ($r = -0.064$), while the r value slightly exceeded the threshold of significance in the case of 'Mv Makaróni' ($r = -0.566$*).

Among the meteorological factors, correlations between the GI and the rainfall data, mean temperatures and number of heat days were analysed using correlation analysis and stepwise regression. Pearson's correlation coefficient indicated that the GI of 'Parus', 'Mv Pennedur' and the mean GI for the three varieties were determined to the greatest extent by the mean temperature in the second ten days of June ($r = -0.709$**, -0.673* and -0.680*, respectively). In the case of the later-maturing variety 'Mv Makaróni' the strongest correlation coefficients were obtained for the rainfall quantity in April and in the last 10 days of June ($r = -0.612$* and -0.556*, respectively). A correlation was also found for 'Parus' with the rainfall quantities in January and March ($r = 0.610$* and 0.679*), and for 'Mv Pennedur' with the number of heat days in the middle 10 days of June ($r = -0.642$*).

Stepwise regression on the mean data of the three varieties and on the data of 'Parus' revealed only one meteorological factor with a significant influence on the GI: this was the mean temperature in the second 10 days of June ($R^2 = 0.462$ and 0.502, respectively). In the case of 'Mv Makaróni' the amount of precipitation during the last 10 days of June, in January and in the second ten days of May had a significant effect on the GI ($R^2 = 0.815$). The GI of 'Mv Pennedur' was also mainly determined by the mean temperature in the second ten days of June, but the number of heat days in the first 10 days of June and the amount of precipitation in the middle 10 days of June also influenced the gluten quality significantly ($R^2 = 0.846$).

Table 5 Significance of the main effects and their interactions tested by Chi squared (F = Wald statistic/d.f.) values in the REML analysis for individual years and the three years combined. Martonvásár, 2010–2012

Source	df	2010	2011	2012	2010–2012
Fertiliser (N)	5	4.00**	3.04	31.93***	3.57**
Fungicide (F)	4	2.98*	0.10	0.36	0.57
Genotype (G)	2	1078.77***	461.386***	272.22***	953.76***
N × F	20	1.12	0.81	1.36	0.55
N × G	10	4.94***	1.24	2.90**	2.01*
F × G	8	1.50	0.59	0.41	0.26
N × F × G	40	0.82	0.41	0.66	0.34
σ^2_{Year}					90.77

Effect of agronomic treatment combinations on the GI of durum wheat varieties

Changes in the GI of three winter durum wheat varieties varying for gluten strength ('Mv Makaróni': weak, 'Mv Hundur': intermediate, 'Mv Pennedur': strong) were investigated in three consecutive years with extreme rainfall supplies (2010–2012) in an experiment involving six nitrogen supply levels and five plant protection treatments. Significant effects were detected for the fertiliser ($F_F = 3.04^*$ − 31.93^{***}) in 2010, 2012 and the 3 years combined (Table 5). The GI averaged over the three varieties increased from 61.83 (0 kg N ha^{-1}) to 67.23 (150 kg N ha^{-1}) in 2010, while it decreased from 60.28 (0 kg N ha^{-1}) to 47.37 (100 + 50 kg N ha^{-1}) in 2012. No significant effect could be observed in 2011. The two counter-moving processes resulted in a slight decrease in the mean values, averaged over the three years (from 0 kg N ha^{-1} = 64.06 to 100 + 50 kg N ha^{-1} = 60.61 kg N ha^{-1}).

The effect of fungicide treatment was statistically proven only in 2010, the year with an excessive amount of precipitation. The GI of samples treated with fungicide both at first node stage and at flowering (treatment 3: 62.380; treatment 5: 62.866) differed significantly from the untreated ones (treatment 1: 66.683; $LSD_{5\%} = 2.809$); however, this difference was practically negligible. The results again proved the primary importance of the genotype (F_G = 272.22^{***} − 1078.77^{***}). A large significant genotypic effect was observed for GI regardless of the wet gluten content (r between wet gluten content and GI was −0.107, −0.203, −0.341 and −0.227 in 2010, 2011, 2012 and the 3 years combined, respectively). A

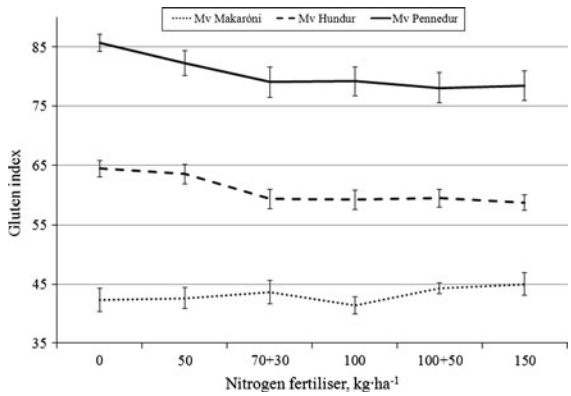

Fig. 2 Effect of nitrogen fertiliser (six levels) on the gluten index of winter durum wheat varieties. Genotype responses represent means of 3 years, five fungicide treatments and three replicates. Martonvásár, 2010–2012

genotype effect was expected, because the samples selected for this study varied greatly in gluten strength. Averaged over years, treatments and replications, the values of the GI for the three varieties differed significantly. The GI averaged 80.45 for 'Mv Pennedur', 60.76 for 'Mv Hundur' and 43.21 for 'Mv Makaróni' ($LSD_{0.1\%} = 1.68$). Despite the substantial difference in the amount of precipitation, the effect of the year was not significant.

The significant fertiliser × genotype interaction indicated that the response to N fertiliser was dependent on the variety. The response of the three varieties to N fertilisation differed in the 3 years. The GI of 'Mv Pennedur' (strong gluten) and 'Mv Hundur' (medium gluten strength) remained stable (in 2010 and 2011) or decreased slightly (in 2012), while that of 'Mv Makaróni' (weak) increased from 28.1 to 47.05 with increasing N levels in 2010. The 3-year average GI

(Fig. 2) of ‘Mv Makaróni’ was the same regardless of the treatment. For ‘Mv Hundur’, which has intermediate gluten strength, the GI of samples originating from the untreated control and from the 50 kg N ha^{-1} treatment was significantly different from that of samples treated with 100 or 150 kg ha^{-1} N active agent. In the case of ‘Mv Pennedur’ a continuous decline in the GI was observed as the N dose increased to a level of 100 kg N ha^{-1}, but no significant difference was observed above this level, regardless of whether the dose was split or not.

Discussion

Effect of genotype on GI

The genotype is the most important factor affecting the gluten strength of durum wheat measured by the GI method. The results of earlier studies (Clarke et al. 2000, 2009) proved that the heritability for this trait was high, ranging between 0.84 and 0.95, and, as recently reported by Longin et al. (2013), the repeatability is also high ($h^2 = 0.90$), even in the case of winter durum wheat genotypes. The present results, performed using different genotypes confirm earlier observations, as the repeatability value averaged over 3 years was 0.93 and was even higher, when the 3 years were treated separately.

The durum wheat varieties and lines included in the study were representative of the history of breeding over the last 30 years. The earliest varieties in the collection were registered in 1980 (‘GK Basa’ and ‘GK Minaret’) and the latest varieties in 2011 (‘Mv Hundur’ and ‘Mv Pennedur’). In the majority of cases, genotypes originating from each individual country (except Russia, represented by only one variety) exhibited a wide range of GI values (Fig. 1).

The mean GI was highest for varieties bred in Germany (70.20), and values of over 85 (the threshold for excellent quality) were recorded for a number of Hungarian, Austrian and Italian durum wheat varieties, averaged over the 3 years.

Weak gluten was characteristic primarily of older, genuinely winter genotypes, probably because gluten strength was not included among the selection criteria when these varieties were developed. Breeders were chiefly concerned with improving adaptability, especially cold tolerance, in addition to which efforts had to be made to raise the yield potential to an acceptable level, since winter durum had to compete with winter wheat in their production zones (Szunics 1986; Dorofeev et al. 1987). Only when these aims had been achieved was it possible to turn attention to improvements in technological quality traits. This process is clearly illustrated by the GI values of winter durum wheat varieties bred in Martonvásár. The Martonvásár durum wheat breeding programme was initiated in 1982, but right up to the mid-1990s selection was only made for kernel type and vitreousness. The use of laboratory equipment for analysis began in 1996, with measurements on the wet gluten content and the GI. The first 4 years of analysis were sufficient to demonstrate that there was great genetic variability for GI in the breeding material and that this technological parameter had little dependence on the year, thus making it possible to carry out efficient selection for stronger gluten. Improvements in the GI had to be achieved while maintaining or improving the values of other traits (cold tolerance, protein content, yellow index). When ‘Mv Makaróni’ was developed, priority was given to yield stability, giving special attention to improving cold tolerance. An excellent level of cold tolerance was successfully combined in this variety with high protein and yellow pigment content, but in most years its gluten strength did not satisfy the criteria raised by the pasta industry. The varieties released in 2011 proved, that it was possible to combine a high level of cold tolerance with good, or even excellent gluten quality.

Effect of meteorological factors on GI

Meteorological factors have a moderate, but nevertheless significant effect on GI. The GI of all three varieties was highest in 2009. In the case of ‘Parus’ and ‘Mv Makaróni’, both of which have weak gluten, the values were outstandingly high in this year, when extremely low wet gluten contents were recorded. The cause of this phenomenon is not yet known, but the wet gluten contents in 2009 were 21.0 % for ‘Parus’, 24.6 % for ‘Mv Makaróni’ and 25.0 % for ‘Mv Pennedur’, compared with average values of 34.29, 36.16 and 36.42 % in the semolina of the three varieties over the 13-year period. It was for this reason that, when testing the gluten strength of early generations, Clarke et al. (2009) concluded that the determination of the GI was more favourable than that of the SDS sedimentation index, as the GI was not dependent on the protein concentration, with which the SDS sedimentation index was in positive correlation. The present results prove that as the wet gluten content approaches an unrealistically low level, there comes a point where a clear negative correlation can be observed between the gluten content and the gluten strength. However, the connection between the GI and the protein/gluten content is somewhat ambiguous. Cubbada et al. (1992) calculated a correlation coefficient of −0.25 with the dry gluten content. The GI was correlated moderately with the wet gluten content (−0.41, Peña 2000) and weakly (−0.32 to −0.25, Clarke et al. 2010) or not significantly (Marchylo et al. 2001) with the protein content. Longin et al. (2013) reported correlation coefficients of 0.38 and 0.45 between the GI and wet gluten content in two sets of winter durum wheat genotypes. These results suggest that the correlation between the wet gluten content and the GI depends on the genotypes tested in the experiments. In the present case the *r* values for the three varieties tested were between −0.295 and −0.566*, but when all three varieties were included in the calculation this negative effect disappeared ($r = -0.064$).

The results of correlation and stepwise regression analysis clearly indicated the primary importance of the temperature during the second 10 days of June for the GI. This period is equivalent to the late-milk to soft-dough stage of grain development for winter durum wheat in Hungary. The fact that heat stress causes changes in the gluten quality of bread wheat has been known for two decades. Blumenthal et al. (1993) detected changes in the ratio of storage proteins, including a shift in the gliadin:glutenin ratio, associated with a softening of the gluten and a deterioration in rheological traits. The unfavourable effect of high temperature during the grain-filling period was also proved in phytotron experiments on Hungarian wheat varieties (Balla et al. 2011). The damaging effect of heat stress on the technological quality, especially the alveograph P/L value, of spring durum wheat was reported by Corbellini et al. (1998), but no data have been found in the literature on the reduction in the GI of winter durum wheat. Further experiments will be required to determine whether the correlation between an increase in the GI of ‘Parus’ and the rainfall sums in January and March could be attributed to a direct effect or whether it was the result of other indirect physiological effects.

It is also clear from the data that varieties with weak gluten exhibited greater variability between years than ‘Mv Pennedur’. Even in the best year the GI of varieties with weak gluten was smaller than the lowest value recorded for ‘Mv Pennedur’, while that of the latter variety exceeded the value of 66 designated by Cubbada et al. (1992) as indicative of ‘good’ gluten quality even in the worst year, and in 9 out of 13 years was higher than the value of 85 required for classification in the ‘excellent’ category.

Effect of agronomy on the GI

Besides the large genotypic effect, the level of nitrogen fertilisation (all years) and fungicide treatments (in 2010) significantly affected the GI of winter durum genotypes. Furthermore the individual durum wheat varieties gave different responses to increasing levels of fertilisation (the N × G interaction was significant in 2010, 2012 and the three years combined). These data confirmed the results of Ames et al. (1999), who found that in diverse environments the GI was determined to an overwhelming extent by the genotype, though the genotype × environment interaction also had a much weaker, but detectable effect.

A significant fertiliser effect was observed in two years and when the values were averaged over three years. These findings contradict those of Ames et al. (2003), who reported that on average, N fertiliser does not significantly influence the gluten strength as measured by GI method. In the present case this statement was only valid in 2011.

The application of fungicide containing azoxystrobin and cyproconazole slightly decreased the GI in 2010. Whether this was a direct or indirect effect of these active agents is not known. The ‘greening effect’ of both strobilurins and triazoles has been well known for more than a decade (Wu and von Tiedemann 2001). These chemicals interfere with the senescence process and elongate the grain-filling period of cereals, suggesting that the plant protection treatment might have a direct physiological effect. However, according to an international patent (Jann et al. 2012) the strobilurins can be used to increase the gluten strength in winter cereals. The indirect effect may be manifested in the suppression or eradication of pathogens affecting the gluten strength. Since the effect of fungicide treatment was practically negligible in a year with unusual weather conditions, this observation is of limited importance.

The varieties respond differently to environmental changes, depending on the gluten type. Varieties with strong or moderately strong gluten are able to maintain a stable gluten structure both in wet and dry years, while in years with excessive precipitation those with weak gluten may produce grain with quality well below the mean value for the variety. However, the combined effect of heat and drought stress may cause the gluten quality of even the strongest genotype to deteriorate. The improvement achieved in the GI by providing different nitrogen supplies was not sufficient in any case to put the variety into a higher quality category.

Conclusion

As in the case of common wheat, the GI is a highly heritable trait in winter durum wheat, with little dependence on the environment. It can be tested on very small samples and provides information on both the quality and quantity of gluten. GI measurements can be successfully used in winter durum breeding programmes for the improvement of gluten strength. Using selection based on this parameter, the gluten strength of winter durum wheat lines can be improved sufficiently to make them competitive with high quality spring varieties.

The demand for high quality durum wheat is increasing worldwide, while the production of this crop species is in decline for various environmental and economic reasons. This contradiction could be resolved partially by expanding the production area of winter durum wheat varieties with good pasta-making quality.

Acknowledgments This research was funded from the grants OTKA K68127 and GOP-1.1.1-09/1-2009-0053. Thanks are due to Dr. Friedrich Longin for his help with the statistics and to an anonymous reviewer, whose suggestions greatly improved the manuscript.

References

Ames NP, Clarke JM, Marchylo BA, Dexter JE, Woods SM (1999) Effect of environment and genotype on durum wheat gluten strength and pasta viscoelasticity. Cereal Chem 76:582–586

Ames NP, Clarke JM, Dexter JE, Woods SM, Selles F, Marchylo BA (2003) Effects of nitrogen fertilizer on protein quantity and gluten strength parameters in durum wheat (*Triticum turgidum* L. var. *durum*) cultivars of variable gluten strength. Cereal Chem 80:203–211

Balla K, Rakszegi M, Li Z, Békés F, Bencze S, Veisz O (2011) Quality of winter wheat in relation to heat and drought shock after anthesis. Czech J Food Sci 29:117–128

Beke B, Barabás Z (1981) The first Hungarian durum wheat varieties. Cereal Res Commun 9:115–117

Blumenthal CS, Barlow EWR, Wrigley CW (1993) Growth environment and wheat quality: the effect of heat stress on dough proprieties and gluten proteins. J Cereal Sci 18:3–21

Bruneel C, Pareyt B, Brijs K, Delcour JA (2010) The impact of the protein network on the pasting and cooking properties of dry pasta products. Food Chem 120:371–378

Clarke JM, Clarke FR, McCaig TN, Knox RE, Ames NP (2000) Evaluation of predictors of quality for use in early generation selection. In: Royo C, Nachit MM, Di Fonzo N, Araus JL (eds) Durum wheat improvement in the Mediterranean region: new challenges. Options Méditerranéennes Série A 40:439–446

Clarke FR, Clarke JM, Pozniak CJ, Knox RE, McCaig TN (2009) Protein concentration inheritance and selection in durum wheat. Can J Plant Sci 89:601–612

Clarke FR, Clarke JM, Ames NA, Knox RE, Ross JR (2010) Gluten index compared with SDS-sedimentation volume for early generation selection for gluten strength in durum wheat. Can J Plant Sci 90:1–11

Corbellini M, Mazza L, Ciaffi M, Lafiandra D, Borghi B (1998) Effect of heat shock during grain filling on protein composition and technological quality of wheat. Euphytica 100:147–154

Cubbada R, Carcea M, Pasqui LA (1992) Suitability of the gluten index method for assessing gluten strength in durum wheat and semolina. Cereal Food World 37:866–869

Cunin C, Handschin S, Walther P, Escher F (1995) Structural changes of starch during cooking of durum wheat pasta. Lebensm Wiss Technol 28:323–328

D'Edigio MG, Mariani BM, Nardi S, Novaro P, Cubbada R (1990) Chemical and technological variables and their relationships: a predictive equation for pasta cooking quality. Cereal Chem 67:275–281

Dexter JE, Matsuo RR (1977) Influence of protein content on some durum wheat quality parameters. Can J Plant Sci 57:717–727

Dexter JE, Matsuo RR (1980) Relationship between durum wheat protein properties and pasta dough rheology and spaghetti cooking quality. J Agr Food Chem 28:899–902

Dorofeev VF, Udachin PA, Semenova LV, Novikova MV, Gradchaninova OD, Shitova IP, Merezhko AF, Filatenko AA (1987) Pshenitsi mira (Wheats of the World). Agropromizdat, Leningrad

Edwards NM, Gianibelli MC, McCaig TN, Clarke JM, Ames NP, Larroque OR, Dexter JE (2007) Relationships between dough strength, polymeric protein quantity and composition for diverse durum wheat genotypes. J Cereal Sci 45:140–149

Feillet P (1984) The biochemical basis of pasta cooking quality, its consequences for durum wheat breeders. Sci Des Aliments 4:551–566

ICC (International Association for Cereal Science and Technology) (1995) ICC Standard No. 158: Gluten index method for assessing gluten strength in durum wheat (*Triticum durum*)

Jann EV, Oerlecke D, Schultz D, Tavares-Rodrigues MA, Zambon S (2012) Use of strobilurins for increasing the gluten strength in winter cereals. BASF, International patent. Publication number: WO/2012/045737, 12 Apr 2012. http://patentscope.wipo.int/search/en/WO2012045737

Lerner SE, Cogliatti M, Ponzio NR, Seghezzo ML, Molfese ER, Rogers WJ (2004) Genetic variation for grain protein components and industrial quality of durum wheat cultivars sown in Argentina. J Cereal Sci 40:161–166

Longin CFH, Sieber AN, Reif JC (2013) Combining frost tolerance, high grain yield and good pasta quality in durum wheat. Plant Breeding 132:353–358

Malcolmson LJ, Matsuo RR, Balshaw R (1993) Textural optimization of spaghetti using response surface methodology: effects of drying temperature and durum protein level. Cereal Chem 70:417–423

Marchylo BA, Dexter JE, Clarke FR, Clarke JM, Preston KR (2001) Relationships among bread-making quality, gluten strength, physical dough properties, and pasta cooking quality for some Canadian durum wheat genotypes. Can J Plant Sci 81:611–620

Matsuo RR, Irvine GN (1967) Macaroni brownness. Cereal Chem 44:78–85

Miedaner T, Longin CFH (2013) Genetic variation for resistance to Fusarium head blight in winter durum material. Crop Pasture Sci. doi:10.1071/CP13170

Palamarchuk A (2005) Selection strategies for traits relevant for winter and facultative durum wheat. In: Royo C, Nachit M, Di Fonzo N, Araus JL, Pfeiffer WH, Slafer GA (eds) Durum wheat breeding: current approaches and future strategies. Food Products Press, New York, pp 599–644

Peña RJ (2000) Durum wheat for pasta and bread-making. Comparison of methods used in breeding to determine gluten quality-related parameters. In: Royo C, Nachit MM, Di Fonzo N, Araus JL (eds) Durum wheat improvement in the Mediterranean region: new challenges. Options Méditerranéennes Série A 40:423–430

Perten H (1990) A rapid measurement of wet gluten quality by the gluten index method. Cereal Food World 35:401–402

Petitot M, Abecassis J, Micard V (2009) Structuring of pasta components during processing: impact on starch and protein digestibility and allergenicity. Trends Food Sci Tech 20:521–532

Sissons M, Abecassis J, Marchylo B, Cubbada R (2012) Methods used to assess and predict quality of durum wheat, semolina and pasta. In: Sissons M, Abecassis J, Marchylo B, Carcea M (eds) Durum wheat: chemistry and technology, 2nd edn. AACC International Inc, St Paul, pp 213–234

Szunics L (1986) A durum búza Magyarországon a hazai szakirodalom tükrében (Durum wheat in Hungary as reflected in domestic special literature). Növénytermelés 35:259–267

Szunics L, Bedő Z, Szunics Lu, Láng L, Veisz O (1998) Results of durum wheat breeding in Martonvásár. Acta Agron Hung 46:135–148

Vasiljevic S, Banasik OJ, Shuey WC (1977) A micro unit for producing durum semolina. Cereal Chem 54:397–404

Virk DS, Pandit DB, Sufian MA, Ahmed F, Siddique MAB, Samad MA, Rahman MM, Islam MM, Ortiz-Ferrara G, Joshi KD, Withcombe JR (2009) REML is an effective analysis for mixed modelling of unbalanced on-farm varietal trials. Expl Agric 45:77–91

Wu YX, von Tiedemann A (2001) Physiological effects of azoxystrobin and epoxiconazole on senescence and the oxidative status of wheat. Pesticide Biochem Physiol 71:1–10