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in

Porceddu E. (ed.), Damania A.B. (ed.), Qualset C.O. (ed.).
Proceedings of the International Symposium on Genetics and breeding of durum wheat

Bari : CIHEAM

Options Méditerranéennes : Série A. Séminaires Méditerranéens; n. 110

2014

pages 583-587

Article available on line / Article disponible en ligne à l'adresse :

<http://om.ciheam.org/article.php?IDPDF=00007120>

To cite this article / Pour citer cet article

Spina A., Ammar K., Peña R.J., Bentivegna G., Sciacca F., Virzi N., Palumbo M. **Durum wheat breeding lines with new HMW glutenin subunit combinations selected for bread-making quality** . In : Porceddu E. (ed.), Damania A.B. (ed.), Qualset C.O. (ed.). *Proceedings of the International Symposium on Genetics and breeding of durum wheat*. Bari : CIHEAM, 2014. p. 583-587 (Options Méditerranéennes : Série A. Séminaires Méditerranéens; n. 110)



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Durum wheat breeding lines with new HMW glutenin subunit combinations selected for bread-making quality

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Abstract. A breeding program was carried out jointly by CIMMYT (Mexico) and CRA-ACM (Italy) in order to improve the bread-making quality of durum wheat (*Triticum turgidum* subsp. *durum*). The research, objective of the research was to expand the genetic diversity for glutenin subunits in durum wheat, and resulted in the development of breeding stocks with new HMW-GS subunits. Ninety-six breeding lines were tested at Sonora (Mexico) in irrigated experimental trials during the 2009-10 cropping season. The same genotypes were grown in Sicily (Italy) in the 2011-12 season under rainfed conditions. Most of the lines showed HMW-GS combinations that included subunits typically found in bread wheat (1 or 2*; 7+17 or 17+18, 5+10 at the Glu-A1, Glu-B1 and Glu-D1 loci, respectively). Main bio-agronomic parameters were evaluated and genotype adaptability in different environments was studied. Gluten quality-related analysis and experimental bread-making test were carried out in order to study the aptitude of semolina with different HMW-GS combinations to bread-making quality. Results showed that the highest bread volumes were obtained from genotypes characterized by the glutenin combination 7+8/5+10 and 7+17/5+10.

Keywords. *Triticum turgidum* subsp. *durum* – Protein composition interspecific crosses – Baking quality.

Lignées de sélection du blé dur avec de nouvelles combinaisons de sous-unités gluténines de haut poids moléculaire (HMW-GS) sélectionnées pour la qualité de panification du pain

Résumé. Un programme de sélection génétique a été réalisé conjointement par le CIMMYT (Mexique) et le CRA-ACM (Italie) afin d'améliorer la qualité de panification du blé dur (*Triticum turgidum* subsp. *durum*). Cette recherche avait pour objectif d'accroître la diversité génétique pour les sous-unités gluténines chez le blé dur, et elle a permis de développer des stocks de reproducteurs avec de nouvelles sous-unités HMW-GS. Quarante-deux lignées ont été testées à Sonora (Mexique) dans des essais expérimentaux effectués avec irrigation au cours de la saison de culture 2009-10. Les mêmes génotypes ont été cultivés en sec, en Sicile (Italie), durant la saison 2011-12. La plupart des lignées ont montré des combinaisons HMW-GS qui comprenaient des sous-unités normalement présentes dans le blé tendre (1 ou 2*, 7+17 ou 17+18, 5+10 au locus Glu-A1, Glu-B1 et Glu-D1, respectivement). Les principaux paramètres bio-agronomiques ont été évalués et l'adaptabilité des génotypes aux différents environnements a été étudiée. Des analyses liées à la qualité du gluten et un essai expérimental de panification ont été réalisés afin d'étudier l'aptitude de la semoule avec différentes combinaisons HMW-GS à la qualité panifiable. Les résultats ont montré que les volumes de pain les plus élevés sont obtenus à partir de génotypes caractérisés par la combinaison de gluténines 7+8/5+10 et 7+17/5+10.

Mots-clés. *Triticum turgidum* subsp. *durum* – Composition en protéines – Croisements interspécifiques – Qualité de cuisson.

I – Introduction

Genetic improvement of durum wheat has always been directed to the enhancement of technological parameters to produce high quality pasta. On the other hand, in many countries a large part of durum wheat production is used for of bread-making and other baked products (Palumbo *et al.*, 2005; Spina *et al.*, 2011a). Worldwide, 76% of durum wheat is used for the production of pasta, while the remaining 24% is used for the production of bread and other baked goods, as well as couscous, bulgur, etc.

In Italy, although most of the durum wheat harvest is devoted to pasta preparation (90.8%), part of durum wheat production is still used for manufacturing local, typical bread, and other baked food forms. UNIPI data for 2009 indicated that 6.6% is used in baking, 2.3% is exported as semolina and 0.3% for other domestic uses.

The bread-making aptitude is influenced especially by gluten extensibility (Ammar *et al.*, 2000; Peña *et al.*, 1994; Liu *et al.*, 1996), which depends on high molecular weight glutenin components (HMW-GS) (Boggini and Pogna, 1989), many of which are present in genotypes derived from interspecific hybridization with *Triticum aestivum*. The presence of genes for those components allows the selection Durum Wheat genotypes characterized by good bread-making quality, irrespective of their origin (Boggini *et al.*, 1995; Spina *et al.*, 2009).

CRA, other institutions in Italy and CIMMYT in Mexico are leading a breeding program to improve bread-making quality of durum wheat (Palumbo *et al.*, 2000; Boggini *et al.*, 2003; Spina *et al.*, 2011b). Durum wheat lines possessing glutenin subunits suitable for bread-making quality were crossed with durum and bread wheat cultivars and lines with good rheological and baking quality were selected from the segregating material. Most of these lines showed HMW-GS combinations that included subunits typically found in bread wheat (1 or 2*; 7+17 or 17+18, 5+10 at the Glu-A1, Glu-B1 and Glu-D1 loci, respectively).

II – Material and methods

Ninety-six F₆BC₅ breeding lines were tested for agronomic traits at Sonora, Yaqui Valley (Mexico), which is 40 m above sea level during the 2009-10 cropping season, in comparison to 4 tester varieties, whereas 99 F₆BC₅ and F₈BC₅ breeding lines (the 96 plus three additional lines) were tested at Libertinia, Sicily (Italy) (180 m above sea level) during the 2011-12 cropping season. Most of the lines, exhibited the HMW 1 and 2* in the A genome; 7+17, 17+18 in the B genome and 5+10 from the D genome, in addition to 6+8 and 7+8 HMW-GS, typical of durum wheat.

A square lattice experimental design, with 10 incomplete blocks and two repetitions was adopted at Sonora; the four rows plots had a 3.36 m² size. Full irrigation was applied. The trial carried out at Libertinia differed from the previous one for the plot size (1.88 m²) and the rainfed conditions. The following bio-morphological parameters were recorded: date of ear emergence and flowering, awns color, and plant vigor. Yield, test weight, and thousand seeds weight were recorded on the harvested material. Electrophoretic analysis of storage proteins was performed on 5 seeds from 5 different spikes from each. Protein content was evaluated by NIR System unit (Foss Tecator) and ISScan software. Additional parameters were recorded on whole grain semolina: yellow index (by Konica Minolta colorimeter CR-410 - Minolta method); sedimentation volume in SDS (using a stirrer apparatus for SDS test) and sedimentation index (as ratio between the sedimentation volume and protein content, as proposed by Peña *et al.*, 1990). Bread-making test was performed, according to the AACC method 10-10. On the grain obtained from trials performed in Italy the following parameters were determined: protein content, gluten content and yellow index (using NIT apparatus - Foss Tecator). Statistical processing of agronomic trials was performed by means the software Genstat (Blues Spatial Analysis Results). The data concerning yield and quality parameters were processed using Statistics software version 6.

III – Results and discussion

Average values for Yield data from Sonora (7.44 t/ha) were higher than those from Libertinia (5.90 t/ha) (Fig. 1). The same trend was observed for grain parameters, such as test weight (TW) (83.2 vs 79.8 kg/hl), and thousand seeds weight (TSW) (47 vs 28.8 g).

Figure 1. Yield results (T_xha⁻¹ of the trials carried out in 2009-10 at Sonora (left) and in 2011-12 at Libertinia (right).

A total of 14 different combinations of HMW-GS were detected in protein electrophoresis analysis (Table 1). D genome HMW subunits were present in both 1A and in 1B chromosomes, e.g. subunits 7+17, 5+10 and 17+18 were present on chromosome 1B, in addition to 6+8 and 7+8 typical of durum wheat germplasm). As far as LMW-GS was concerned, all lines were previously selected for the presence of type 2 subunits, and did not show any difference among them.

Table 1. High molecular weight glutenin subunit (HMW-GS) combinations and their frequency in the genotypes analyzed.

Lines with the same HMW-GS No.	HMW-GS	
	1A glu-A1	1B glu-B1
16	Null	7+8
14	Null	7+17
10	Null	6+8
8	Null	17+18
8	6+8	5+10
8	7+17	5+10
6	2*	17+18
6	7+8	5+10
6	17+18	5+10
4	1	7+8
4	2*	7+17
2	1	6+8
2	1	7+17
2	2*	7+8

Protein content was significantly higher in the Libertinia harvest (16.1%, range 15.2 - 17.0%) than that from Sonora (12.0%, range 10.0 - 14.0%) (Fig. 2). The sedimentation volume of whole grain semolina was on average more than 12 cm³ in the Sonora material, confirming the good quality of the gluten of most genotypes. Gluten content had an average value of 11.8% and a range of variation between 10.8 and 12.5%.

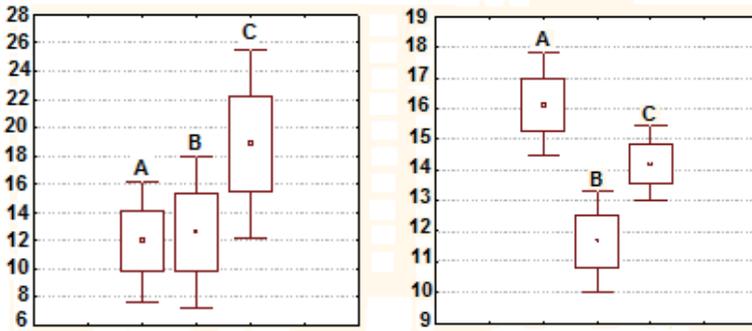


Figure 2. Grain protein (A), sedimentation volume (B) and grain color (C) in the analyzed material grown at Sonora (left) and at Libertinia (right).

Yellow index exhibited significant variability (from 15 to 22 b*) and satisfactory average value (19 b*), in the material from Sonora, whereas the material from Libertinia had showed lower average value (14.2 b*) and narrow range of variability.

The results of experimental baking test carried out on the 2010 trial are showed in table 2. The selected lines showed 557.1 cm³ average value of bread volume, higher than that of durum wheat testers. The bread wheat testers, as expected, reached the highest volumes (> 800 cm³).

Bread volume was significantly correlated with the sedimentation index ($r = - 308^{**}$).

Table 2. Results of baking test.

Bread-making parameters	Lines			Durum wheat Testers			Common wheat testers		
	Mean	Range	St. Dev.	Mean	Range	St. Dev.	Mean	Range	St. Dev.
Volume (cm ³)	557.1	445-785	63.9	504.4	475-650	81.3	807.1	740-900	56.6
Porosity (1-5)*	3.8	2-5	0.9	2.9	1-5	1.4	1,4	1-2	0.53
Crumb color (b**)	30.7	24.5-42.6	2.1	27.2	22.1-31,9	4.2	15.1	12.6-17.3	1.8

Concerning the relation between the glutenin composition and bread volume, the genotypes with the protein combinations 7+8/5+10, 7+17/5+10, 17+18/5+10 and 2*/17+18 showed higher bread volume (about 600 cm³), indicating good bread-making quality. The 7+8/5+10, 7+17/5+10 HMW-GS combinations showed the best bread volume (616.7 and 616.3 cm³, respectively).

The results of the study indicated that the introgression and expression of genes from bread wheat into durum wheat can be an efficient strategy for improving the bread-making quality of tetraploid wheat where such need exists for cultural, traditional, or local preferences.

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