Theor Appl Genet (2001) 102:767–772

L.A. Pflüger · L.M. Martín · J.B. Alvarez

Variation in the HMW and LMW glutenin subunits from Spanish accessions of emmer wheat (*Triticum turgidum* ssp. *dicoccum* Schrank)

Received: 13 June 2000 / Accepted: 3 July 2000

Abstract Emmer wheat (*Triticum turgidum* ssp. *dicoccum* Schrank) is hulled wheat that survives in marginal areas of the Mediterranean Region. The HMW and LMW glutenin subunit composition of 97 accessions of emmer wheat from Spain have been analysed by SDS-PAGE. For the HMW glutenin subunits, four allelic variants were detected for the *Glu-A1* locus; one of them has not been previously described. For the *Glu-B1* locus, three of the nine alleles detected have not been found before. A high degree of variation was evident for the LMW glutenin subunits, and up to 23 different patterns were detected for the B-LMW glutenin subunits. Considering both types of proteins (HMW and LMW), 30 combinations were found between all the evaluated lines. This wide polymorphism can be used to transfer new quality genes to wheat, and to widen its genetic basis.

Keywords Genetic diversity · Hulled wheats · Glutenins

Introduction

Emmer wheat (*Triticum turgidum* ssp. *dicoccum* Schrank), a primitive hulled wheat, is an allopolyploid species with the genome formula **AABB**, which was widely cultivated in the past under the name of 'farrum'. Currently, this species survives in marginal farming areas of Italy, the Balkan peninsula and Turkey, where it is mainly used both for feeding livestock and for human consumption (Hammer and Perrino 1984; D'Antuono 1989; Galterio et al. 1994). In Spain, it is still possible to find small pockets of hulled wheat cultivation which have survived in isolated areas associated with traditional agriculture and, in some cases, with very archaic practices (Peña-Chocarro 1995). In Asturias, where it is known as 'escanda', 'escaña' or 'povia', emmer has been grown until very recently in several areas; however, today its pure crop cultivation is limited to the central part of the province, and is usually mixed with spelt (*T. aestivum* ssp. *spelta* L. em. Thell.).

The revival of traditional food has increased the interest in hulled wheats over the last few years. This is due to the low-input techniques used for their management (D'Antuono 1989), the increasing demand for unconventional foods, and the therapeutic properties attributed to their derivatives (Auricchio et al. 1982). Some authors have indicated that the quality characteristics of emmer wheat are very poor compared with durum and bread wheats (Galterio et al 1994; Piergiovanni et al 1996; Piergiovanni and Blanco 1999). Nevertheless, it is important to emphasise that the ever-narrowing quality specifications of modern industries should not be overestimated when evaluating the potential of this crop. The potential of emmer wheat does not reside in its capacity to mimic attributes that are already present in the commercial cultivars of wheat, but rather in its genetic potential to obtain new products with high digestibility and non-toxicity for coeliac disease. In addition, the variability of emmer wheat could be a useful gene reservoir for the breeding programmes of durum and bread wheats (Sharma et al. 1981; Srivastava and Damania 1989).

Studies carried out have indicated that the analysis of seed storage proteins (glutenins and gliadins) is a useful tool for quality improvement, because these proteins have proved to be important for the technological properties of wheat. Although the genetic diversity of gliadins and glutenins within emmer germplasm is still under investigation, some analyses carried out in this species have shown considerable allelic variation for high-molecular-weight (HMW) glutenin subunits coded at the *Glu-1* loci (Vallega and Waines 1987; Blanco et al. 1991; Galterio et al. 1994; Piergiovanni and Blanco 1999).

Communicated by H.F. Linskens

L.A. Pflüger · L.M. Martín · J.B. Alvarez (✉)
Departamento de Genética,
Escuela Técnica Superior de Ingenieros Agrónomos y de Montes,
Universidad de Córdoba, Apdo. 3048, E-14080 Córdoba, Spain
e-mail: ge2alcaj@uco.es

Present address:
L.A. Pflüger, Instituto de Recursos Biológicos, CIRN, INTA Castelar,
Las Cabañas y los Reseros s/n, (1712) Castelar. Argentina

Table 1 Accessions of *T. turgidum* ssp. *dicoccum* used in this study

Accessions	No.	Received from
BG-01926, BG-01927, BG-01946, BG-01958, BG-01960, BG-01962, BG-01966, BG-01974, BG-01989, BG-01993, BG-01995, BG-01999, BG-12298, BG-12299, BG-12301, BG-12302, BG-12304, BG-12305, BG-12306, BG-12308, BG-12310, BG-12312, BG-12313, BG-12314, BG-12315, BG-12316, BG-12317, BG-12318, BG-12319, BG-12320, BG-12321, BG-13620, BG-13640, BG-13641, BG-13643, BG-13644, BG-14255, BG-18906, BG-19261, BG-25420	36	Bank of Germplasm. CRF-INIA (Spain)
PI-190920, PI-190921, PI-190922, PI-190923, PI-190924, PI-190925, PI-190926, PI-190927, PI-190931, PI-191091, PI-254193, PI-256031, PI-275996, PI-275997, PI-275998, PI-275999, PI-276000, PI-276001, PI-276002, PI-276003, PI-276004, PI-276005, PI-276006, PI-276007, PI-276008, PI-276009, PI-276010, PI-276011, PI-276012, PI-276013, PI-276014, PI-276015, PI-276016, PI-276017, PI-276018, PI-276019, PI-276020, PI-276021, PI-276022, PI-277012, PI-277670, PI-277671, PI-277672, PI-277674, PI-277675, PI-277676, PI-277677, PI-277678, PI-277681, PI-308879, PI-348620, PI-352336, PI-352337, PI-352338, PI-352342, PI-352344, PI-352346, PI-355482, PI-355483, PI-355484, PI-355485, PI-355486	61	National Small Grain Collections. USDA-ARS. Aberdeen (Idaho, USA)

Because of the possibilities it offers for the detection of novel variants, and for the study of the evolution and domestication of wheat, a wide screening of this primitive germplasm could be relevant. Likewise, the knowledge of its characteristics is fundamental for its utilisation in breeding programmes aimed at improving the performance of this primitive crop, as well as for using it as a source of useful genes for other cultivated wheats.

The main goal of this study was to evaluate the polymorphism of HMW and LMW glutenin subunits present in a collection of emmer wheat from Spain.

Materials and methods

Plant material

Ninety seven emmer accessions, obtained from Centro de Recursos Fitogenéticos INIA (Alcalá de Henares, Spain) and the National Small Grain Collections (Aberdeen, USA), were analysed in this study (Table 1). Seeds of the HMW glutenin subunit standards (Payne and Lawrence 1983; Vallega and Waines 1987) were obtained from diverse sources. The LMW glutenin subunit standards (Nieto-Taladriz et al. 1997) were kindly provided by Dr. J.M. Carrillo (ETSIA, Univ. Pol. Madrid, Spain).

Protein extraction

Seeds crushed into a fine powder were used to extract the endosperm storage proteins. Gliadins were extracted with a 1.5 M dimethylformamide aqueous solution in a 1:5 ratio (mg/µl) and stored at –20°C. The pellet was double-washed with 50% (v/v) propan-1-ol at 60°C for 30 min, with agitation every 10 min. Glutenin was extracted with 250 µl of buffer containing 50% (v/v) propan-1-ol, 80 mM Tris-HCl pH 8.5, and 2% (w/v) dithiothreitol at 60°C for 30 min. After centrifugation, 200 µl of the supernatant were transferred to a new tube, mixed with 3 µl of 4-vinylpyridine, and incubated for 30 min at 60°C. The samples were precipitated with 1 ml of cold acetone. The dried pellet was solubilized in buffer containing 625 mM Tris-HCl pH 6.8, 2% (w/v) SDS, 10% (v/v) glycerol, 0.02% (w/v) bromophenol blue, and 2% (w/v) dithiothreitol in a 1:5 ratio (mg/µl) to wholemeal.

Electrophoretic methods

Glutenins were analysed in vertical SDS-PAGE slabs in a discontinuous Tris-HCl-SDS buffer system (pH 6.8/8.8) at a polyacrylamide concentration of 8% (w/v, C: 1.28%) for the study of HMW glutenin subunits. To elucidate some allelic variants, a further analysis by SDS-PAGE was performed, using 8% gels containing 4 M urea as reported by Lafiandra et al. (1993). LMW glutenin subunits were analysed by 10% gels with 4 M urea. Electrophoresis was performed at a constant current of 35 mA/gel at 18°C for 45 min after the tracking dye (bromophenol blue) migrated off the gel.

Results and discussion

In recent times, numerous studies have been performed to evaluate the materials conserved in germplasm banks with molecular markers as new tools. In the case of wheat, these tools have been used for the search of variability at the level of endosperm storage-proteins because of its association with bread-making quality. This search has been performed between the old varieties and landraces of the present or past cultivated wheats. Unfortunately, many of the materials conserved in germplasm banks are only mirrors of old collections with low reliable, or scarce, geographical and agronomic data.

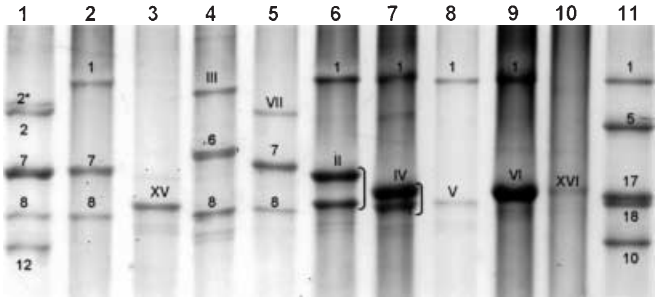

Fig. 1 SDS-PAGE (8%) patterns of HMW-Gs from some Spanish accessions of emmer wheat, representative of the different allelic variants detected at the *Glu-A1* and *Glu-B1* loci. *Lanes*: *1* Buck Charrua; *2* BG-01946; *3* BG-012302 ; *4* PI-190924; *5* PI-308879; *6* BG-012301; *7* PI-275996; *8* BG-001989; *9* PI-190931; *10* PI-277681; and *11* Yecora

Most of the materials evaluated in this work, conserved in two germplasm banks, are not present geographical data and only the common names used in the collected zone are available. On the other hand, as numerous institutional or private collectors have shown, this has complicated the evaluation of possible duplications of the materials. Therefore, we have evaluated the present collection as independent accessions, because no data on agronomic or other characteristics are available. Obviously, it is possible that accessions with different names belong to the same landrace or vice versa.

Novel variants of HMW glutenin subunits

In Fig. 1, the HMW glutenin subunit composition of the several emmer accessions, representative of the variation detected, is shown. Up to 13 allelic variants (four alleles at the *Glu-A1* locus and nine at the *Glu-B1* locus) were found in the evaluated accessions (Table 2). Four novel allelic variants were identified, one at the *Glu-A1* locus and three at the *Glu-B1* locus. Although the International nomenclature indicated by McIntosh et al. (1998) has been used to name the alleles previously described, including the alleles found in emmer wheat by Vallega and Waines (1987), we have considered it more convenient to name the new alleles with the progressive Roman-numeral nomenclature of Vallega and Waines (1987) and Branlard et al. (1989). These new alleles were designated *Glu-A1-VII* and *Glu-B1-XV*, *Glu-B1-XVI* and *Glu-B1-XVII*. The subunits from the alleles found by Vallega and Waines (1987) were named with the Roman number used by these authors for the designation of the alleles, independently of whether these alleles were formed by one or two components.

Similar to other cultivated wheats (Shewry et al. 1992), only one active component was found for the *Glu-A1* locus. Between the alleles previously described in wheat (Payne and Lawrence 1983), the allele *Glu-A1a* (subunit 1) was most frequent among the evaluated accessions (86 of 97), while the null allele (*Glu-A1c*) was observed in only seven accessions (Table 2). These results show a sharp discrepancy with those of Vallega and Waines (1987), who found that the null allele appeared with a frequency of 21% in 167 accessions from the 23 countries analysed.

Table 2 Comparison of allele frequencies for *Glu-A1* and *Glu-B1* loci among 97 Spanish accessions of emmer wheat and among 167 accession of the world collection

Locus	Allele	Subunits	This work		Vallega and Waines[a]	
			No.	%	No.	%
Glu-A1	a	1	86	89	93	56
	c	Null	7	7	35	21
	j	III (one)	3	3	12	7
	VII	VII (one)	1	1	–	–
Glu-B1	b	7+8	71	73	18	11
	d	6+8	5	5	30	18
	n	II (two)	9	9	15	9
	p	IV (two)	1	1	22	13
	q	V (one)	1	1	3	2
	r	VI (one)	1	1	22	13
	XV	XV (one)	7	7	–	–
	XVI	XVI (one)	1	1	–	–
	XVII	XVII (two)	1	1	–	–

[a] Vallega and Waines (1987)

The allele *Glu-A1j*, described as *Glu-A1-III* by Vallega and Waines (1987) for emmer, was found in three accessions (PI-190924, PI-277675 and PI-348620); however, none of these accessions carry the alleles *Glu-A1h* (subunit I) and *Glu-A1i* (subunit II) that were found in the world collection analysed by the same authors. A novel allele, *Glu-A1-VII* (Fig. 1, lane 5), that was only found in one accession (PI-308879) only showed a band with a very fast mobility, being even faster than subunit 2* coded by the *Glu-A1b* allele (Fig. 1, lane 1), and which was not found among the accessions analysed in this report.

For the *Glu-B1* locus, the allele *Glu-B1b* (subunits 7+8) was most frequent among the evaluated lines (73%), contrary to what was observed in the collection analysed by Vallega and Waines (1987), where these subunits were found in only 11% of the accessions. The allele *Glu-B1d* (subunits 6+8) was present in five accessions.

The allele *Glu-B1o* (subunit III), which was very common in the world collection analysed by Vallega and Waines (1987), was not found in the present work, neither was the *Glu-B1m* allele (subunit I). The allele *Glu-B1n* (subunit II), formed by two major components, was present in nine accessions (9%), a frequency similar to that reported by Vallega and Waines (1987). The subunits IV (allele *Glu-B1p*), V (allele *Glu-B1q*) and VI (allele *Glu-B1r*) were each found in only one accession, respectively.

The new allele *Glu-B1-XV* presents only one major component, which was slightly faster than the subunit VI coded by *Glu-B1r* and was found in seven accessions. On the other hand, only one accession (PI-277681) has the new allele

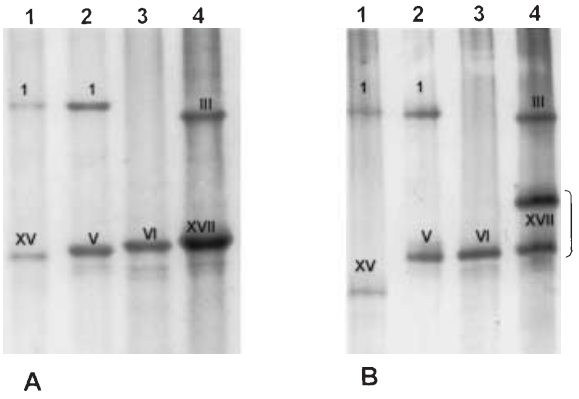

Fig. 2 SDS-PAGE, without (**A**) and with (**B**) 4 M urea, of some allelic variants detected at the *Glu-B1* locus. *Lanes*: *1* PI-190922; *2* PI-094633 (*Glu-B1q* control); *3* PI-094669 (*Glu-B1r* control); *4* PI-348620

Glu-B1-XVI, which showed only one band, with a slightly slower mobility than subunit VI (Fig. 1, lanes 9 and 10).

Although the SDS-PAGE technique is usually performed by separation of the glutenin subunits, diverse studies have shown that it is possible that these proteins present anomalous mobilities (Goldsbrough et al. 1989, Lafiandra et al. 1993). The cause of this behaviour seems to be due to conformational differences and this anomalous mobility can be eliminated by the addition of a strong denaturant, such as 4 M urea, to the gel (Goldsbrough et al. 1989). In the present work, this technique has been used to detect possible new variants.

When urea was added to gel, the mobility of all subunits showed changes. Sometimes, this has better differentiated some subunits. For example, the subunit coded by the *Glu-B1-XV* allele (Fig. 2A, lane 1), that was slightly faster-moving than subunit V of *Glu-B1q* (Fig. 2A, lane 2), was faster in urea gels (Fig. 2B, lanes 1 and 2, respectively). Furthermore, one new allele was found. The allele present in the PI-348620 accession was first identified as the *Glu-B1r* allele (Fig. 2A, lane 4). Nevertheless, when urea was applied to the gel, this allele showed a new pattern formed by two components with very different mobility (Fig. 2B, lane 4). This new allele was named *Glu-B1-XVII*.

Considering the variation for the *Glu-A1* and *Glu-B1* loci, 13 patterns were detected. Their frequencies are shown in Table 3. It is to be emphasised that 31 of the 36 accessions obtained from CRF-INIA (Spain) showed the same composition (A*x*1, B*x*7+B*y*8), which was also the most frequent (47.5%) among the accessions from the NGSC (USA). This last collection also showed a higher degree of variation than the first one. Twelve patterns were found among the NGSC accessions, but nine of them were not found in the CRF-INIA collection. For the CRF-INIA accession, only four patterns were detected; the composition A*x*1+B*x*V present in accession BG-001989 was not found in the NGSC collection.

Table 3 Frequencies of the HMW glutenin subunit compositions among 97 Spanish emmer accessions

Origins	Subunit composition		No.	%	Accession standards
	1A	1B			
CRF-INIA (Spain) no.=36	Null	XV	3	8.30	–
	1	7+8	31	86.10	–
	1	II	1	2.80	BG-012301
	1	V	1	2.80	BG-001989
NGSC (USA) no.=61	Null	II	1	1.60	PI-277674
	Null	XV	2	3.30	–
	Null	XVI	1	1.60	PI-277681
	1	6+8	3	4.90	–
	1	7+8	29	47.50	–
	1	II	7	11.50	–
	1	IV	1	1.60	PI-275996
	1	VI	1	1.60	PI-190931
	1	XV	2	3.30	–
	III	6+8	2	3.30	–
	III	XVII	1	1.60	PI-348620
	VII	7+8	1	1.60	PI-308879

Variations in LMW glutenin subunits

Liu and Shepherd (1996), using 19 lines of two collections of emmer wheat from at least six countries, found that the B-LMW glutenin subunits present a high degree of variation. In fact, they detected up to 16 different patterns with 4–6 bands, varying in mobility and staining intensity. In the present work, we have used a SDS-PAGE gel with 4 M urea, which has allowed us to detect greater separation between the B-LMW glutenin subunits. Some electrophoretic migration patterns can be seen in Fig. 3A and B.

In Fig. 3C, a diagram of the 23 different patterns found in these lines is shown. These patterns are formed by 2–6 B subunits coded at the *Glu-A3* and *Glu-B3* loci, the patterns with four or five bands being the most frequent (9 and 8 of 23, respectively). The differences between the patterns were for mobility and staining intensity. In fact, some patterns could be understood simply as variants. For example, the differences between the patterns 1, 2, 7 and 14 are only at the position of the first band, similar to that observed between the patterns 3, 4, 17, 19 and 21 (Fig. 3C). The relative frequencies of each pattern are shown in Table 4. The most-frequent patterns were 4, 5 and 6, which were found in 61 of the evaluated lines (62.9%). By contrast, 12 patterns were only found in one line. The rest appear to be distributed between seven patterns.

When the HMW and LMW glutenin subunits were simultaneously studied, up to 30 different combinations were found; however, only four of them were common to both collections (Table 5). Twenty one combinations appear only between accessions from the NSGC, and five were detected in the CRF-INIA collection. It is important to emphasize that the three common combinations (A*x*1, B*x*7+B*y*8, LMW-4; A*x*1, B*x*7+B*y*8, LMW-5; and A*x*1, B*x*7+B*y*8, LMW-6) represent 59.8% of all the eval-

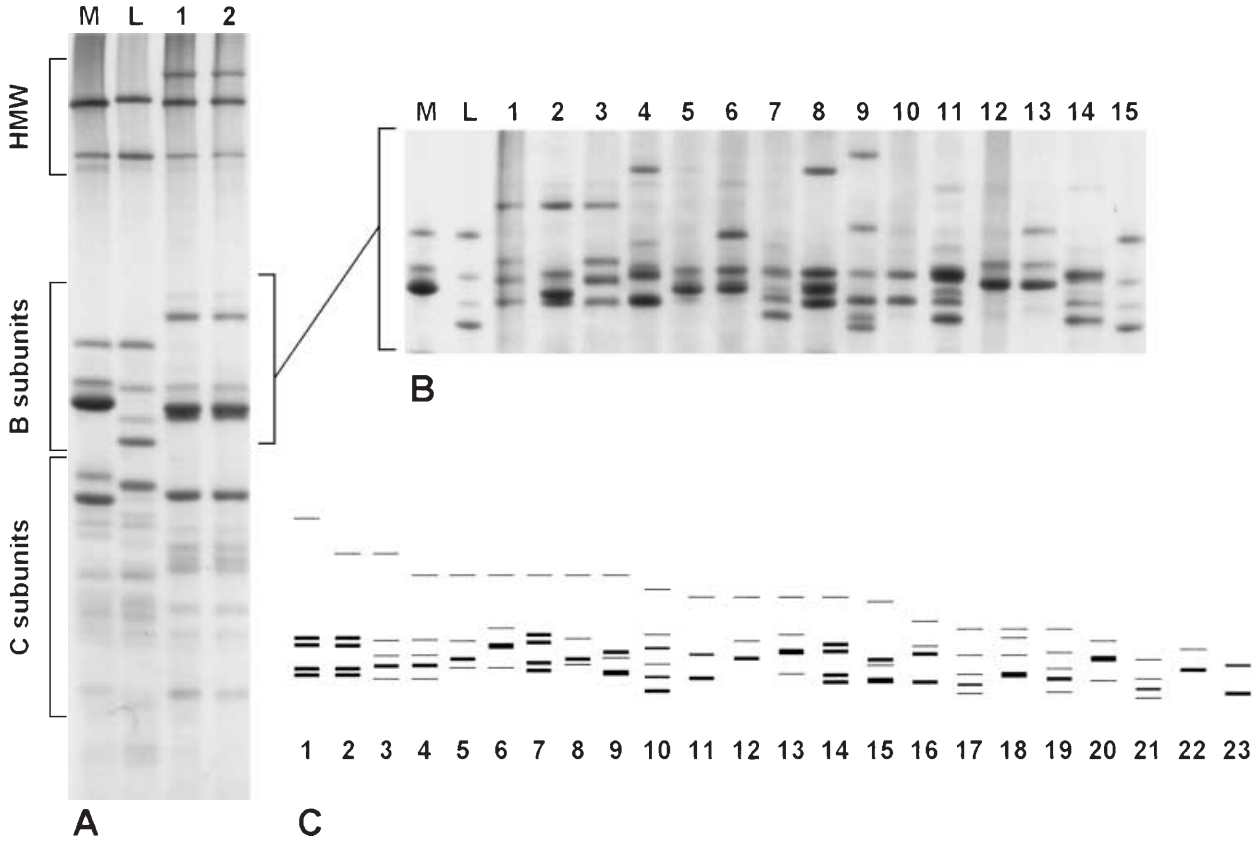

Fig. 3A–C SDS-PAGE (10% with 4 M urea) separation of LMW glutenin subunits from some Spanish accessions. **A** HMW and LMW glutenin subunits (*M* 'Mexicali', standard LMW-2; *L* 'Langdon', standard LMW-1); **B** only the lower part of the gel corresponding to the LMW-Gs is shown; **C** diagram of all patterns found for B-LMW glutenin subunits

Table 4 Frequencies of various LMW glutenin subunit compositions among 97 Spanish accessions of *T. turgidum* ssp. *dicoccum*

Patterns	N	%
1	1	1.0
2	2	2.1
3	1	1.0
4	20	20.6
5	19	19.6
6	22	22.7
7	1	1.0
8	5	5.2
9	4	4.1
10	1	1.0
11	3	3.1
12	3	3.1
13	1	1.0
14	1	1.0
15	2	2.1
16	1	1.0
17	1	1.0
18	1	1.0
19	1	1.0
20	1	1.0
21	3	3.1
22	1	1.0
23	2	2.1

uated accessions (75% in the CRF-INIA collection, and 51% in the NSGC collection).

Conclusions

In normal SDS gels, the relative mobility of some novel subunits detected amongst Spanish accessions in the present work is only slightly different from those of certain variants described by Vallega and Waines (1987), and their presence might have been overlooked in their research. In fact, we have been able to detect such novel allelic variants as *Glu-B1-XV* and *Glu-B1-XVII* with the aid of urea gels. In normal 8% gels, these subunits have nearly the same mobility as subunit VI (*Glu-B1r*).

In spite of having found high allelic variability for the LMW glutenin subunits, the presence of the same pattern for HMW glutenin subunits in many accessions (A*x*1, B*x*7+B*y*8) was noteworthy. However, considering the entire banding pattern, most of them were different for the LMW glutenin subunits. It would be interesting to make a quality evaluation of these materials, considering that the results of Galterio et al. (1998) showed that emmer lines with this allelic composition (A*x*1, B*x*7+B*y*8) had the highest sedimentation volume. It is also relevant to evaluate the quality of those genotypes that carried new subunits.

On the other hand, besides new alleles not previously described, the information presented may also be of interest to plant breeders for choosing parents to obtain re-

Table 5 Frequencies of the HMW/LMW glutenin subunit compositions among 97 Spanish emmer accessions

Subunit composition		CRF-INIA (no.=36)	NGSC (no.=61)	Total (no.=97)	
HMW	LMW	No.	No.	No.	%
1, 6+8	17	–	1	1	1.0
	20	–	1	1	1.0
	21	–	1	1	1.0
1, 7+8	3	–	1	1	1.0
	4	13	7	20	20.6
	5	6	10	16	16.5
	6	8	14	22	22.7
	8	2	–	2	2.1
	9	1	3	4	4.1
	13	–	1	1	1.0
	18	1	–	1	1.0
	19	–	1	1	1.0
	21	–	2	2	2.1
1, II	2	–	1	1	1.0
	7	1	–	1	1.0
	11	–	1	1	1.0
	12	–	1	1	1.0
	16	–	1	1	1.0
1, IV	10	–	1	1	1.0
1, V	5	1	–	1	1.0
1, VI	23	–	1	1	1.0
1, XV	15	–	2	2	2.1
III, 6+8	22	–	1	1	1.0
	23	–	1	1	1.0
III, XVII	14	–	1	1	1.0
Null, II	2	–	1	1	1.0
Null, XV	5	–	2	2	2.1
	8	3	–	3	3.1
Null, XVI	12	–	1	1	1.0
VII, 7+8	1	–	1	1	1.0

combinant lines with good bread-making quality. The wide polymorphism detected should be evaluated for its effects on technological properties through the transfer of new allelic variants to durum wheat. We think that this species could be used as a source of genes for quality improvement in durum wheat.

Acknowledgements This research was supported by grant AGF98–0945-C02–02 from the Spanish Interministerial Commission of Science and Technology (CICYT). We thank the Bank of Germplasm INIA (CFR, Alcalá de Henares, Spain) and the National Small Grain Collection (Aberdeen, USA) for supplying the emmer wheat accessions, and Dr. J.M. Carrillo (ETSIA, Univ. Pol. Madrid, Spain) for standard LMW wheats. L.A. Pflüger is grateful to the Spanish Agency of International Cooperation (AECI) for a predoctoral fellowship.

References

Auricchio S, De Ritis G, De Vincenzi M, Occorsio P, Silano V (1982) Effects of gliadin-derived peptides from bread and durum wheat on small-intestine culture from rat fetus and coeliac children. Pediatr Res 16: 1004–1010

Blanco A, Giorgi B, Perrino P, Simeone R (1991) Risorse genetiche e miglioramento della qualità del frumento duro. Agricoltura e Ricerca 144:41–58

Branlard G, Autran JC, Monneveux P (1989) High-molecular-weight glutenin subunits in durum wheat (*T. durum*). Theor Appl Genet 78:353–358

D'Antuono LF (1989) Il farro; areali di coltivaziones, caractteristiche agronomiche, utilizzazione e prospettive colturali. L'Informatore Agrario 45:49–57

Galterio G, Cappelloni M, Desiderio E, Pogna NE (1994) Genetic, technological and nutritional characteristics of three Italian populations of "farrum" (*Triticum turgidum* ssp. *dicoccum*) J Genet Breed 48:391–398

Galterio G, Codianni P, Novembre G, Saponaro C, Di Fonzo N, Pogna NE (1998) Storage-protein composition and technological characteristics of F_6 lines from the cross *Triticum turgidum* ssp. *durum*×*Triticum turgidum* ssp. *dicoccum*. In: Slinkard AE(ed) Proc 9th Int Wheat Genet Symp, vol. 4. University Extension Press, University of Saskatchewan, Saskatoon, Canada, pp 148–150

Goldsbrough AP, Bulleid NJ, Freedman RB, Flavell RB (1989) Conformational differences between two wheat (*Triticum aestivum*) 'high–molecular–weight' glutenin subunits are due to a short region containing six amino acid differences. Biochem J 263:837–842

Hammer K, Perrino P (1984) Further information on farro (*Triticum monococcum* L. and *Triticum dicoccum* Schrank) in South Italy. Kulturpflanze 32:143–151

Lafiandra D, D'Ovidio R, Porceddu E, Margiotta B, Colaprico G (1993) New data supporting high Mr glutenin subunit 5 as a determinant of qualitative differences in the pairs 5+10 vs 2+12. J Cereal Sci 18:197–205

Liu CY, Shepherd KW (1996) Variation of B subunits of glutenin in durum, wild and less-widely cultivated tetraploid wheats. Plant Breed 115:172–178

McIntosh RA, Hart GE, Devos KM, Gale MD, Rogers WJ (1998) Catalogue of gene symbols for wheat. In: Proc. 9th Int. Wheat Genet. Symp, vol 5. University Extension Press. University of Saskatchewan. p 235

Nieto-Taladriz MT, Ruiz M, Martinez MC, Vazquez JF, Carrillo JM (1997) Variation and classification of B low–molecular–weight glutenin subunit alleles in durum wheat. Theor Appl Genet 95:1155–1160

Payne PI, Lawrence GJ (1983) Catalogue of alleles for the complex gene loci, *Glu–A1*, *Glu–B1* and *Glu–D1*, which code for high–molecular–weight subunits of glutenin in hexaploid wheat. Cereal Res Comm 11:29–35

Peña-Chocarro L (1995) *In situ* conservation of hulled wheat species: the case of Spain. In: Padulosi S, Hammer K, Heller J (eds) Hulled wheats. IPGRI, Rome, Italy, pp 130–148

Piergiovanni AR, Blanco A (1999) Variation of HMW glutenin and γ-gliadin subunits in selected accessions of *Triticum dicoccum* (Schrank) and *T. spelta* (L.) Cereal Res Commun 27:205–211

Piergiovanni AR, Laghetti G, Perrino P. (1996) Characteristics of meal from hulled wheats (*Triticum dicoccum* Schrank and *T. spelta* L.): an evaluation of selected accessions. Cereal Chem 73:732–735

Sharma HC, Waines JC, Foster KW (1981) Variability in primitive and wild wheats for useful genetic characters. Crop Sci 21: 555–559

Shewry PR, Halford NG, Tatham AS (1992) High-molecular-weight subunits of wheat glutenin. J Cereal Sci 15:105–120

Srivastava JP, Damania AB (1989) Use of collections for cereal improvement in semi-arid areas. In: Brown AHD, Frankel OH, Marshall DR, Williams JT (eds) The use of plant genetic resources. Cambridge University Press, UK, pp 88–104

Vallega V, Waines JG (1987) High-molecular-weight glutenin subunit variation in *Triticum turgidum* var. *dicoccum*. Theor Appl Genet 74:706–710