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Genetic diversity of European spelt wheat (*Triticum aestivum* ssp. *spelta* L. em. Thell.) revealed by glutenin subunit variations at the *Glu-1* and *Glu-3* loci

Xueli An[1,‡], Qiaoyun Li[1,‡], Yueming Yan[1,*], Yinghua Xiao[1], S.L.K. Hsam[2] & F.J. Zeller[2,*]
[1]*Key Laboratory of Genetics and Biotechnology, College of Life Science, Capital Normal University, 100037 Beijing, China;* [2]*Division of Plant Breeding and Applied Genetics, Technical University of Munich, D85350 Freising-Weihenstephan, Germany*
*(*authors for correspondence: e-mail: yanym@hotmail.com, zeller@zwz.tum.de)*

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Summary

High and low molecular weight glutenin subunit (HMW-GS and LMW-GS) compositions of 270 European spelts, 15 Iranian spelts and 25 bread wheat cultivars were analyzed by one- and two-dimensional gel electrophoresis. The results revealed a total of 22 HMW-GS alleles (4 at *Glu-A1*, 11 at *Glu-B1* and 7 at *Glu-D1*) and 32 allele combinations among the three *Glu-1* loci. Two major genotypes of HMW-GS: 1, 13+16, 2+12 and 1, 6.1+22.1, 2+12 were found to occur in Central European spelt wheat cultivars and landraces at higher frequencies of 35 and 28%, respectively. The *Glu-B1* locus displayed the greatest variation and genetic diversity index (H) was 0.69 whereas *Glu-A1* and *Glu-D1* showed H index values of 0.26 and 0.19, respectively. The dendrogram constructed by HMW and LMW glutenin subunit bands revealed that European spelts form a separated cluster from common wheat suggesting that spelt and common wheat form distinct groups. In addition, all 15 Iranian spelt land variety accessions differed from European spelts and possessed similar HMW-GS alleles to common wheat. Because of a wider polymorphism Central European spelt wheats are an important genetic reserviour for improving common wheat quality.

Introduction

Spelt wheat (*Triticum aestivum* ssp. *spelta* L. em. Thell, 2n = 6x = 42, AABBDD) was one of the major feed and food grains in ancient Europe. Compared with common wheat, spelts possess some typical morphological characteristics: a narrow, lax and pyramidal spike with a brittle rachis and adherent glumes, generally long spike internodes and non-spherical seeds (Mac-Key, 1966; Campbell, 1997). Although spelt wheat is now considered a minor crop, renewed interest has been developing in recent years because of the increasing demand for unconventional foods and low-input agriculture, its outstanding stress resistance, and quality performance (Campbell, 1997). Previous investigations showed that spelt wheat possessed many useful genes, including resistance to yellow rust (Kema, 1992; Campell, 1997), stem rust, leaf rust; dwarf bunt, Septoria nodorum blotch, and barley yellow dwarf virus, as well as excellent bread-making quality, higher vitamin content and higher nutrition values (Campbell, 1997).

Wheat storage proteins, including glutenins and gliadins are the main components of gluten and glutenins subgrouped into high and low molecular weight glutenin subunits (HMW and LMW-GS), encoded by the *Glu-1* (*Glu-A1*, *Glu-B1* and *Glu-D1*) and *Glu-3* (*Glu-A3*, *Glu-B3* and *Glu-D1*) sets of loci, respectively (Payne, 1987; Shewry, 1992). It has been well documented that glutenins have major effects on bread-making quality due to their contributions to dough strength (Payne, 1987). In addition, storage proteins are also reliable genetic markers for investigating genetic diversity in wheat (Branlard et al., 2001; Abdel-Aal et al., 1996), marker-assisted selection

[‡]Both authors contributed equally to this work.

(Ahmad, 2000) and wheat origin and evolution (Yan et al., 2003a).

In the past, some effort has been made to improve biotic stress resistance, specifically for resistance to various rusts and Septoria tritici blotch, as well as grain quality by hybridization between spelt and common wheat (Kema, 1992; Campbell, 1997). The genetic variability of spelt collections appears to have potential use for wheat improvement. In this study, we use the improved one- and two-dimensional polyacrylamide gel electrophoresis (PAGE) methods to determine the allelic compositions of HMW and LMW glutenin subunits and genetic diversity of European spelts.

Materials and methods

Plant materials

The materials analyzed included 270 spelt cultivars and lines (*Triticum aestivum* L. ssp. *spelta* L.) mainly originating from Central European countries, and 15 primative Iranian spelt landraces. These were kindly provided by the genebanks of Braunschweig and Gatersleben, Germany, and the Plant Germplasm Institute, Kyoto University, Japan. Twenty five common wheat cultivars, including 18 European cultivars (Apostle, Brimstone, Longbow, Avalon, Beaver, Riband, Alpe 1, Floreal, Liocorno, Ilves, Nisu, Gemini, Timone, Ruso, Pandas, Pricama, Prinqual and Salmone) kindly provided by Dr. G. Branlard, France. These 18 cultivars have been used as standards for HMW and LMW glutenin subunit identification as recommended by Jackson et al. (1996).

One- and two-dimensional polyacrylamide gel electrophoresis

Improved procedures for HMW and LMW glutenin subunit extraction and SDS-PAGE (Yan et al., 2003b) were used. A-PAGE of HMW-GS was performed with a Boi-Rad Mini-PROTEAN 3 cell according to Yan et al. (1999). Two-dimensional A-PAGE x SDS-PAGE was carried out according to Redaelli et al. (1995) with some modifications. The first-dimensional A-PAGE was carried out at 500V for 25min, and then the gels were cut into single strips and incubated for 20min at room temperature in the equilibration solution containing 10% glycerol, 2% SDS, 0.0625M Tris-HCl at pH6.8. The equilibrated gel strips were placed on the top of the second-dimension SDS-PAGE gel (12%). The second-dimensional electrophoresis was carried out in the same apparatus as described above. Electrophoresis was performed at 12mA for 2h, and then gels were stained by the routine method.

Allele identification

The identifications of high and low molecular weight glutenin subunits were based on the classification of Payne and Lawrence (1983) and Jackson et al. (1996), respectively. The new subunits and alleles identified were designated according to Yan et al. (2003a) and McIntosh et al. (2003).

Data analysis

The genetic diversity at each locus was calculated on the basis of Nei (1973): $H = 1 - \Sigma\ {p_i}^2$ (H is Nei's genetic variation index and p_i is the frequency of a particular allele at that locus). For cluster analysis, data were scored on the presence or absence of HMW and LMW glutenin bands. The presence of a particular glutenin band in a accession was assigned a value of 1 whereas absence was designated by 0. The data matrices obtained were analyzed with SIMQUAD (similarity for qualitative data) routine of NYSYS-PC to generate Jaccard similarity coefficients, which were used to construct a dendrogram using the unweighted pair group method with arithmetic averages (UPGMA) by SHAN clustering routine from NTSYS (Abdel-Aal et al., 1996).

Results and discussion

Alleleic composition of HMW glutenin subunits

According to a previous study (Yan et al., 2003b), both HMW and LMW glutenin subunits can be well separated by the improved one- and two-dimensional PAGE methods. In this work, two types of glutenin subunits of spelt and common wheat cultivars were separated and characterized by SDS-PAGE and by A-PAGE × SDS-PAGE. The compositions and frequencies of HMW-GS and some typical LMW-GS are shown in Table 1. Of 270 European spelt cultivars and accessions analyzed, a total of 22 allelic variants at three *Glu-1* loci were detected. In comparison with the previous investigations on Spanish spelt accessions (Rodriguez-Quijano et al., 1990; Caballero et al., 2001), eight novel

Table 1. Allelic composition of HMW- and LMW glutenin subunits at different loci in 270 European spelt accessions

			Accessions		Previous reports (%)*	
Locus	Allele	Subunit	No.	%	A	B
Glu-A1	*a*	1	229	84.81	86.60	84.70
	b	2*	5	0.85	12.41	2.60
	c	null	34	12.59	0.99	12.70
	w	2.1*	2	0.74	–	–
	H			0.26		
Glu-B1	*a*	7	2	0.74	–	–
	b	7 + 8	39	14.44	–	–
	c	7 + 9	3	1.11	–	–
	d	6 + 8	1	0.37	–	–
	f	13 + 16	112	41.48	87.84	87.30
	be	6.1 + 22.1	91	33.70	–	–
	bf	6.1 + null	3	1.11	–	–
	bg	13* + 19*	7	2.59	–	–
	bh	13 + 22*	10	3.70	–	–
	bi	13 + 22.1	1	0.37	–	–
	bj	14* + 15*	1	0.37	–	–
	H			0.69		
Glu-D1	*a*	2 + 12	243	90.00	89.08	89.90
	b	3 + 12	8	2.96	8.68	3.30
	c	4 + 12	1	0.37	–	–
	d	5 + 10	11	4.07	0.25	–
	e	2 + 10	4	1.48	–	–
	l	null + 12	2	0.74	0.50	4.20
	bp	2.1' + 12	1	0.37	–	–
	H			0.19		
					C	
Glu-A3	*a*		87	32.22	51.02	
	h		165	61.11	–	
Glu-B3	*d*		188	69.63	4.08	
	f		22	8.15	8.16	
	g		41	15.19	28.57	
Glu-D3	*a*		166	61.48	59.18	
	c		104	38.52	40.82	

*A. Caballero et al. (2001). B. Rodriguez-Quijano et al. (1990). C. Results from 49 European bread cultivars by Jackson et al. (1996). H: Nei's index.

alleles (*w*, *be*, *bf*, *bg*, *bh*, *bi*, *bj*, *bp*) and nine new subunits of HMW-GS were found. Some representative HMW-GS and LMW-GS alleles are shown in Figure 1 (SDS-PAGE) and Figure 2 (2-DE).

At the *Glu-A1* locus, only four HMW-GS alleles were detected. Alleles *a* (subunit 1) and *c* (null) were present at frequencies of 84.48% and 12.59%, respectively. Five accessions possessed allele *b* (subunit 2*) at a frequency of 0.84%. A novel subunit designated 2.1* (allele *w*) was located between subunits 1 and 2* in two Spanish spelt accessions (1026 and 1094). This result is different from a previous report on Spanish spelt accessions by Caballero et al. (2001), who found that alleles *b* and *c* were present at frequencies of 12.41% and 0.99%, respectively.

At the *Glu-B1* locus, 11 HMW-GS alleles were detected and six were novel (7 new subunits). Two alleles, *Glu-B1f* (13 + 16) and *Glu-B1be* (6.1 + 22.1) were the most frequent, and detected in 41.48% and 33.7%, respectively of the accessions. The HMW-GS 13 + 16 were considered to have positive effects on bread-making quality and had the same quality score as subunits 17 + 18 and 7 + 8 at the *Glu-B1* locus (Gianibelli et al., 2001). As in the reports of Caballero et al. (2001) and Rodriguez-Quijano et al. (1990), the allele *Glu-B1f* occurred frequently in Spanish spelt accessions whereas it was rare in common wheat (Payne & Lawrence, 1983). However, Central European spelt cultivars mainly originating from Germany and Switzerland contained the novel allele *Glu-B1be* (6.1 + 22.1) at much higher frequency (Table 1). As shown in Figure 1, subunit 6.1 moves slightly faster than subunit 6 and subunit 22.1 has similar mobility to subunit 18. Another allele appearing at a higher frequency (14.44%) was *Glu-B1b* (7 + 8). This allele, as well as the other three alleles, occurred at lower frequencies; *Glu-B1a* (subunit 7), *Glu-B1c* (7 + 9) and *Glu-B1d* (6 + 8) were not previously detected in Spanish spelt collections (Rodriguez-Quijano et al., 1990; Caballero et al., 2001), but occurred in common wheat at higher frequencies (Payne & Lawrence, 1983). In addition, five new rare alleles designated *bg* (13* + 19*), *bh* (13 + 22*), *bf* (6.1 + null), *bi* (13 + 22.1) and *bj* (14* + 15*) were found in European spelts. Six spelt cultivars and lines: Rechenbergs Früher Dinkel, Renval, Zeiners Weißer Schlegel and KU-3410 from Germany, TRI9885/74 from Belgium and accession SP1 from China possessed the subunit pair 13* + 19*. Their mobilities were distinct from that of the subunit 13. Ten accessions possessed the allele *bh* (13 + 22*) with the novel subunit 1By22* (=chromosome 1B, subunit y 22*) situated between 1By22.1 and 1By8. Three accessions KU-3418, KU-3446 and TRI 4613/75 from Germany possessed only the 1Bx6.1 subunit, whereas the 1By gene was silent. The 13 + 22.1 subunit pair was present in accession KU-1135 from Spain, while TRI11553/92 from Russia contained 14* + 15* subunits, located between subunits 14 and 15.

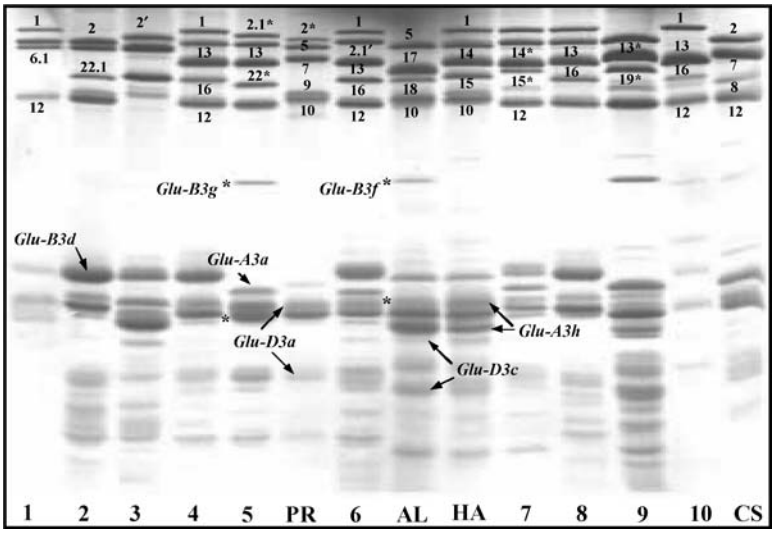

Figure 1. SDS-PAGE patterns of 10 European spelt wheat cultivars and accessions showing representative HMW and LMW glutenin subunits. (1) KU-3418, (2) Steiners Roter Tiroler, (3) TRI14165/91 (cultivated emmer), (4) KU-1135, (5) KU-1094, (6) KU-1034, (7) TRI11553/92, (8) Bauländer Spelz, (9) SP1, (10) KU-1137. Common wheat: PR Pricama, AL- Alidos, HA Hanno, CS Chinese Spring. Some typical subunits and alleles are indicated.

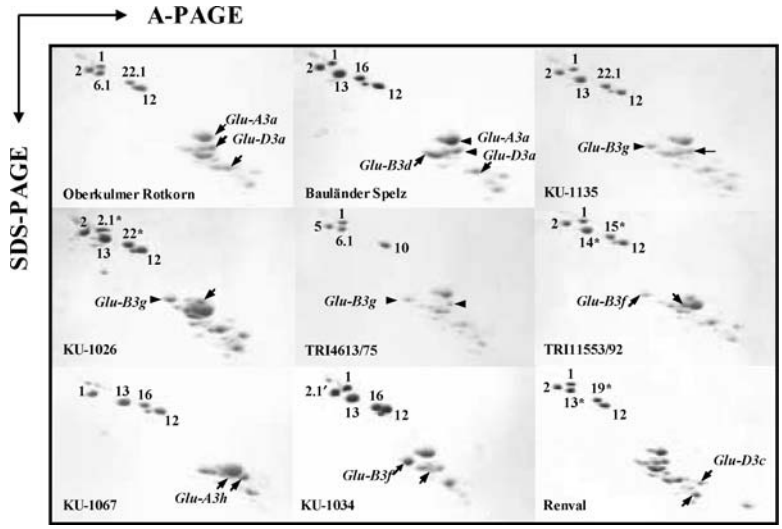

Figure 2. Two-dimensional electrophoresis (A-PAGE × SDS-PAGE) of HMW and LMW glutenin subunits from nine Central European spelt wheats. Some typical alleles are indicated.

Seven alleles including a new allele named *Glu-D1bp* (2.1' + 12) were found at the *Glu-D1* locus. As shown in Figure 1 (lane 7, no.6) the novel subunit designated 2.1', was situated between 2* and 2. The cultivated emmer *Triticum turgidum* ssp. *dicoccum* accession TRI14165/91 was also found to have a subunit (designated 2') with the same mobility (lane 3, Figure 1) but encoded by the *Glu-A1* locus. The allele *Glu-D1a* (2 + 12) was the most frequent (90%) among the materials analyzed whereas the better quality subunit pair 5 + 10 was present only in seven accessions (4.07%). Two accessions (KU-1137 and KU-1067) originating from Spain contained only a y-type subunit 1Dy12 (allele *Glu-D1l*). The remaining three alleles *Glu-D1b* (3 + 12), *Glu-D1c* (4 + 12) and *Glu-D1e* (2 + 10) were detected at lower frequencies. *Glu-D1c*

Table 2. Frequencies of HMW glutenin subunit compositions among 270 European spelt accessions

HMW-GS compositions			Alleles	Number of accessions	Frequencies (%)	Accession sample
Glu-A1	*Glu-B1*	*Glu-D1*				
1	13 + 16	2 + 12	*a, f, a*	95	35.19	Bauländer Spelz
2*	13 + 16	2 + 12	*b, f, a*	1	0.37	Sp158
N	13 + 16	2 + 12	*c, f, a*	1	0.37	KU-3430
1	13 + 16	3 + 12	*a, f, b*	6	2.22	TRI2128/75
1	13 + 16	4 + 12	*a, f, c*	1	0.74	KU-1110
1	13 + 16	5 + 10	*a, f, d*	1	0.37	KU-1131
1	13 + 16	2.1' + 12	*a, f, bp*	1	0.37	KU-1034
1	13 + 16	null + 12	*a, f, l*	2	0.74	KU-1067
1	13 + 16	2 + 10	*a, f, e*	4	1.48	KU-1078
2.1*	13 + 22*	2 + 12	*w, bh, a*	2	0.74	KU-1026
1	13 + 22*	2 + 12	*a,bh, a*	6	2.22	KU-1086
1	13 + 22*	3 + 12	*a,bh, b*	1	0.37	Grado
N	13 + 22*	2 + 12	*c,bh, a*	1	0.37	KU-1139
1	13 + 22.1	2 + 12	*a,bi, a*	1	0.37	KU-1135
N	6.1 + 22.1	2 + 12	*c, be, a*	8	2.96	Hercule
1	6.1 + 22.1	2 + 12	*a,be, a*	75	27.78	Schwabenkorn
1	6.1 + 22.1	3 + 12	*a, be, b*	1	0.37	SP3
1	6.1 + 22.1	5 + 10	*a, be, d*	7	2.59	Rouguin
1	6.1 + null	5 + 10	*a, bf, d*	1	0.37	TRI4613/75
1	6.1 + null	2 + 12	*a,bf, a*	2	0.74	KU-3418
N	7	2 + 12	*c, a, a*	1	0.37	TRI5648/91
N	7	5 + 10	*c, a, d*	1	0.37	NGB4798.1
N	7 + 8	2 + 12	*c, b, a*	15	5.56	Erbeweizen
2*	7 + 8	2 + 12	*b, b, a*	4	1.48	2678–1563
N	7 + 8	5 + 10	*c, b, d*	1	0.37	TRI13157/91
1	7 + 8	2 + 12	*a, b, a*	19	7.04	TRI474/75
1	13* + 19*	2 + 12	*a,bg, a*	3	1.11	Renval
N	13* + 19*	2 + 12	*c, bg, a*	4	1.48	KU-3410
N	6 + 8	2 + 12	*c, d, a*	1	0.37	TRI9870/82
1	7 + 9	2 + 12	*a, c, a*	2	0.74	TRI13351/95
N	7 + 9	2 + 12	*c, c, a*	1	0.37	TRI5609/84
1	14* + 15*	2 + 12	*a,bj, a*	1	0.37	TRI11553/92

and *Glu-D1e* were not found in the Spanish spelt accessions reported by Rodriguez-Quijano et al. (1990) and Caballero et al. (2001).

Frequencies of HMW-GS compositions at the three loci among 270 European spelt accessions are shown in Table 2. A total of 32 combinations was detected at the *Glu-A1*, *Glu-B1* and *Glu-D1* loci. Two genotypes (1, 13 + 16, 2 + 12 and 1, 6.1 + 22.1, 2 + 12) were the most frequent among all accessions, with 35.19% and 27.78% respectively. Two other combinations: 1, 7 + 8, 2 + 12 (7.04%) and N, 7 + 8, 2 + 12 (5.56%) appeared in low frequencies with the remaining combinations occurring rarely. The 13 Iranian spelt accessions showed common alleles and allele combinations found in common wheat. Two alleles, *Glu-A1b* and *Glu-A1c*, at the *Glu-A1* locus and three alleles, *Glu-B1a*, *Glu-B1b* and *Glu-B1c*, at the *Glu-B1* locus were found. All 13 accessions possessed the allele *Glu-D1a*. Among the three HMW-GS loci, only four allele combinations, *cba*, *bba*, *cca* and *caa* were detected.

Allelic composition of LMW subunits at the Glu-3 loci

Seven alleles at the *Glu-3* loci were identified in European spelts (Table 1). A new allele at *Glu-A3* locus previously identified by Yan et al. (2003a) and

designated *Glu-A3h*, was present in the European spelts at a frequency of 61.11% (indicated in Figure 1 and Figure 2). Another allele, *Glu-B3d*, occurred at a very high frequency (69.63%) in spelts, but was present (4.08%) at low frequency in European common wheat cultivars (Jackson et al., 1996). The frequencies of *Glu-A3a* (31.22%) and *Glu-B3g* (15.19%) in spelt wheats were much lower than those in European common wheat (51.02% and 28.57%, respectively). The *Glu-D3* alleles, *Glu-D3a*, *Glu-D3c* and *Glu-B3f* showed similar frequencies in both spelt and common wheat (Table 1).

Previous investigations showed that some HMW-GS changed their mobilities when separated by A-PAGE (Morel, 1994; Yan et al., 2003b). In the present study, the accessions with novel glutenin subunits were further analyzed by A-PAGE × SDS-PAGE 2D-electrophoresis. Both HMW-GS and LMW-GS were well separated and identified. As shown in Figure 2, contrary to SDS-PAGE, the subunits 1 and 2.1* as well as 2* (not shown) moved faster than subunit 2, and subunit 10 moved faster than subunit 12 on the A-PAGE gel. Additionally, the subunits 13* and 19* showed faster mobilities than subunits 13 and 16, respectively. For the remaining subunits, mobility ranks were similar to that with SDS-PAGE. Results from IEF × SDS-PAGE (Holt et al., 1981) indicated that subunit 12 in most wheat materials was separated into two components: a major and a minor dot. Analysis by A-PAGE × SDS-PAGE in the present study showed the same finding for subunit 12, whereas most of HMW-GS showed a single component. Some typical *Glu-3* alleles identified based on Jackson et al. (1996) occurred in European spelt wheats at a higher frequency and were found to encode between 1 and 3 protein components (Figure 2).

Genetic diversity analysis of HMW glutenin subuinit variation

Nei's genetic variation index (H) was calculated at each HMW-GS locus (Table 1). The *Glu-B1* locus displayed a much higher genetic diversity (0.69) than the *Glu-D1* and *Glu-A1* loci, showing H values of 0.19 and 0.26, respectively. The mean genetic diversity index was 0.38. For the 13 Iranian spelt accessions, the genetic variability of HMW-GS was rather low and H values at *Glu-A1*, *Glu-B1* and *Glu-D1* loci were 0.42, 0.27 and 0, respectively. The 25 common wheat cultivars showed higher genetic diversity although no new alleles at the three *Glu-1* loci were detected. At the *Glu-A1*, *Glu-B1* and *Glu-D1* loci, three (*a*, *b* and *c*), six (*a, b, c, d, h* and *i*) and three (*a*, *b* and *d*) alleles were found and genetic diversity indices were 0.64, 0.81 and 0.53, respectively.

Cluster analysis

In order to determine the genetic relationship between European spelt and common wheat, 70 European spelt, 2 Iranian spelt and 25 common wheat cultivars were used for cluster analysis. The European spelt subgroup was selected so that all the lines with different HMW and LMW-GS alleles and allele combinations, as well as spelts from different localities, were included. The HMW-GS of Central European spelts contained 1, 6.1 + 22.1 or 13* + 19*, 2 + 12; accessions from Spain had 1, 13 + 16, 2 + 12; and Iranian spelts were N, 7 + 8, 2 + 12. The LMW-GS patterns within each group, especially those from Spain, exhibited low variability. The dendrogram constructed on the basis of HMW-GS and LMW-GS is shown in Figure 3, where it is evident that spelt was separated from common wheat. Spelt and the common wheats, except for Chinese Spring (CS), were clustered into two different groups. As shown in Figure 3, the lower hierarchy clusters were generally in agreement with their origins.

Two subclusters (BS-SC and ZD-KU3409) from Central European countries (Germany, Belgium and Austria) include cultivars and lines with HMW-GS compositions mainly including 13 + 16, 13* + 19* and 6.1 + 22.1 at the *Glu-B1* locus. Three subclusters (OR-AR, KU9884-DU and KU3420-3434) involved spelt wheats, mainly from Germany normally possessing the HMW-GS composition: 1, 6.1 + 22.1 and 2 + 12. Both Oberkulmer Rotkorn (OR) and *T. aestivum* L. ssp. *spelta* var. *duhamelianum* (DU) were Central European spelt landraces and therefore could represent the primitive spelt wheat type. The glutenin subunit patterns of two Iranian spelt wheats (KU 3377 and DV1130) were the same as that of Chinese Spring and grouped into the same subcluster.

In the past few decades, introgression of spelt germplasm in wheat improvement has been increasing (Campbell, 1997). Thus, it is important to understand the genetic diversity of the available spelt collections as well as the relationship to common wheat. Storage proteins of wheat endosperm have been shown to be reliable markers not only in the genetic improvement of bread-making quality (Payne, 1987), but also in the studies of crop origin and evolution (Fernandez-Calvin & Orellana, 1990; Yan et al., 2003a). As shown in this study, although limited variation at the *Glu-1* loci was detected in European spelt wheats, genetic diversity of

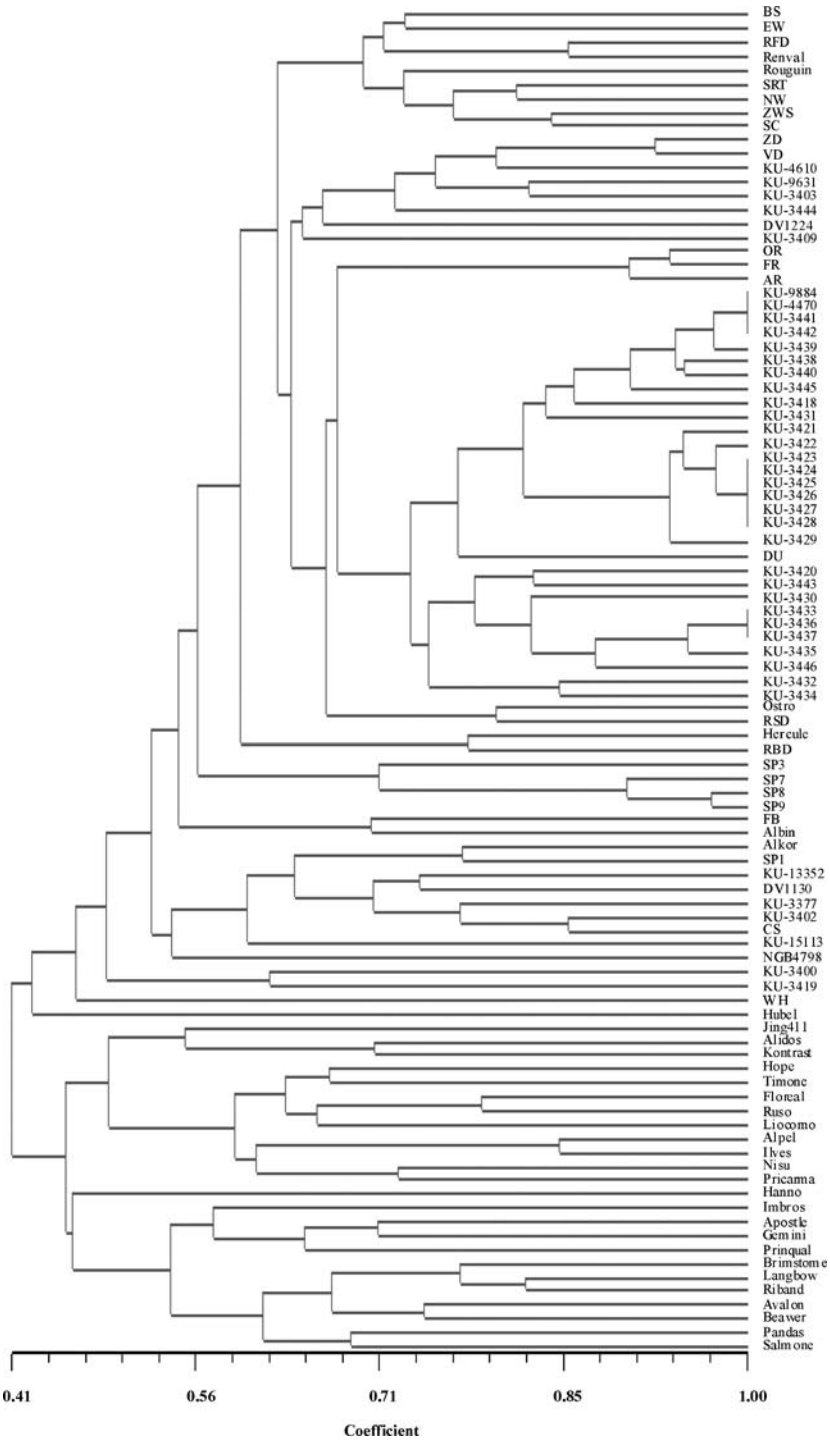

Figure 3. Dendrogram of HMW and LMW glutenin subunit compositions of 70 representative European spelt cultivars and accessions, 2 Iran spelt accessions (KU-3377 and DV1130), 23 European bread wheat cultivars and 2 Chinese common wheat cultivars (CS and Jing 411). The scale indicates the similarity coefficient. (BS) Bauländer Spelz, (EW) Erbe-Weizen, (RFD) Rechenbergs Früher Dinkel, (SRT) Steiners Roter Tiroler, (NW) Neuegger Weißkorn, (ZWS) Zeiners Weißer Schlegel, (SC) Schwabenkorn, (ZD) Zuzger Dinkel, (VD) Vöglers Dinkel, (OR) Oberkulmer Rotkorn, (FR) Franckenkorn, (AR) Altgolder Rotkorn, (DU) *T. aestivum* var. *duhamelianum*, (RSD) Roter Schlegel Dinkel, (RBD) Rechenbergs Brauner Dinkel, (FB) Fuggern Babenhausener, (CS) Chinese Spring, (WH) Waggershauser Hohenheimer.

HMW-GS was higher than previously reported among Spanish spelt accessions (Rodriguez-Quijano et al., 1990; Caballero et al., 2001). A total of 22 alleles, including nine novel subunits and eight new alleles at *Glu-1* loci, and 32 subunit combinations were found. In particular, the *Glu-B1* locus possessed 11 alleles (six novel) and showed the highest genetic diversity index (up to 0.69). Cluster analysis showed that spelt and common wheats were separated into two groups suggesting that significant differences in HMW and LMW-GS compositions exist in these two groups. A previous survey showed that spelt-specific HMW alleles were present in modern and ancient European spelts (Blatter et al., 2002). Results obtained from RFLP and microsatellite molecular markers also revealed a distinct genetic distance (GD) between spelt and common wheat although relatively low GD values were present among European cultivated spelts (Siedler et al., 1994; Bertin et al., 2001). Some investigations indicated high polymorphisms between spelt and common wheat in gliadin patterns (Harsch et al., 1997) and the presence of a spelt-specific γ-gliadin gene (von Büren et al., 2000). Additionally, spelt has more favorable traits than common wheat in many aspects, including stress tolerance and high nutritional value (Luo et al., 2000), supporting the earlier suggestions by Mac-Key (1954) that the difference between spelt and common wheat cannot be explained by just a few genes.

The present study shows that Central European spelt wheat mainly originating from Germany and Switzerland possesses two common alleles at the *Glu-B1* locus, namely *f* (13 + 16, 41.48%) and *be* (6.1 + 22.1, 33.7%). Additionally, ten and seven spelt accessions from this area possessed alleles *bh* (13 + 22*) and *bg* (13* + 19*), respectively. Furthermore, several rare alleles, *w* (2.1*), *bf* (6.1 + null), *bi* (13 + 22.1), *bj* (14* + 15*) and *bp* (2.1' + 12) were also found at three *Glu-1* loci. These novel alleles may be useful for wheat quality improvement. As reported earlier (Yan et al., 2003a) most of the alleles described above, as well as some of the LMW-GS alleles, were also found in club wheat (*T. aestivum* ssp. *compactum* (Host) MK) and cultivated emmer (*T. turgidum* ssp. *dicoccum* (Schrank) Thell.). This strongly supported the hypothesis that European spelt wheat originated from hybridization between cultivated emmer and club wheat (Mac-Key, 1966; Ohtsuka, 1998). Our results also showed that the HMW-GS patterns of primitive Iranian spelt wheat were more similar to common wheat than to European spelt. However, differences existed in the frequencies of y-type alleles of *Glu-B1-1* and *Glu-A1-2* between Asian spelt and common wheat (Blatter et al., 2004). Thus depending on the characters of interest both European and Asian spelts serve as potential germplasm for the improvement of common wheat.

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