ISSN 1479-2621
Plant Genetic Resources **4**(2); 134–143
DOI: 10.1079/PGR2006110

Composition of and variation in high- and low-molecular weight glutenin subunits, and omega gliadins in Ethiopian tetraploid wheat germplasm

Faris Hailu[1]*, Eva Johansson[2], Arnulf Merker[2], Getachew Belay[3], Harjit-Singh[4] and Habtamu Zeleke[4]

[1]*University of Lackomelza, Office of PIC, P.O. Box 33809, Addis Ababa, Ethiopia,* [2]*Department of Crop Science, Swedish University of Agricultural Sciences, P.O. Box 44, SE-230 53 Alnarp, Sweden,*[3]*Debre Zeit Agricultural Research Centre, P.O. Box 32, Debre Zeit, Ethiopia and* [4]*Department of Plant Science, Alemaya University, P.O. Box 219, Alemaya, Ethiopia*

Received 6 September 2005; Accepted 9 January 2006

Abstract

A collection of 120 Ethiopian tetraploid wheat accessions was analysed for high-molecular weight (HMW) glutenin subunit, low-molecular weight (LMW) glutenin subunit and omega gliadin composition by SDS–PAGE. For the HMW glutenin subunits, a new allelic variant, 2****, was detected which has not been previously described at the Glu-A1 locus. A high proportion of Glu-A1x banding pattern was observed in durum wheat. For the Glu-B1 locus four different banding patterns were detected. Among those HMW glutenin subunits, 7 + 8 were the most common, while subunits 14 + 15 and 6 + 8 were found to be rare. A high degree of variation was evident for the LMW glutenin subunits and D-zone omega gliadins. The association of the composition of the gluten with quality has been discussed. This wide variation can be used in improving the quality of wheat and to widen its genetic base.

Keywords: accessions; Ethiopia; high-molecular weight glutenin subunits; low-molecular weight glutenin subunits; omega gliadins; quality of tetraploid wheat

Introduction

Durum wheat (*Triticum durum* Desf.) is well known for superior pasta and macaroni products compared to other wheat species because of its kernel size, hardiness and golden amber colour. An essential element of pasta cooking quality is the ability of the protein components to interact during pasta processing, resulting in insoluble aggregates and visco-elastic complexes entrapping starch granules and limiting surface disintegration of pasta during cooking. Visco-elasticity of cooked pasta and macaroni correlates with protein content and type (Damidaux *et al.*, 1980; Kosmolak *et al.*, 1980; Galterio *et al.*, 1993).

Gluten protein composition in tetraploid wheat can be used for determination of the quality of the wheat. The major endosperm storage proteins in wheat are the polymeric glutenins and the monomeric gliadins, and these proteins can hydrate together forming a cohesive, elastic three-dimensional gluten network. Glutenin is composed of high-molecular weight (HMW) (80–120 kDa) and low-molecular weight (LMW) (30–60 kDa) subunits (Payne and Corfield, 1979), while gliadins are classified into alpha, beta, gamma and omega gliadins based on their electrophoretic mobility at low pH (Woychik *et al.*, 1961). The omega gliadins are also known as D-zone gliadins. Durum wheat has been classified according to the pattern of LMW and HMW glutenins, as well as

* Corresponding author. E-mail: Markhmets@yahoo.com

gamma and omega gliadins (Pogna *et al*., 1988; Kovacs *et al*., 1995; Liu and Shepherd, 1995; Liu and Rathjen, 1996). Quality of durum wheat can be determined on the basis of the presence of gamma gliadin 45 for good, and gamma gliadin 42 for poor pasta quality (Kosmolak *et al*., 1980). The effects of gamma gliadins 42 and 45 are due to their genetic linkage with LMW glutenin subunits designated LMW-1 and LMW-2, respectively (Payne *et al*., 1984).

The HMW glutenin subunits are encoded by genes on the long arm of homoeologous chromosomes-1 (Glu-A1, and Glu-B1 in tetraploid wheat) whereas the genes encoding the LMW subunits are clustered on the short arm of the same chromosome group (Glu-A3, and Glu-B3) (Beitz *et al*., 1975; Jackson *et al*., 1983; Payne *et al*., 1984; Shewry *et al*., 1986). Gliadins are encoded by the Gli-1 and Gli-2 loci located on the distal part of the short arm of the homoeologous group 1 and group 6 chromosomes (Metakovsky *et al*., 1984). The genes encoding LMW glutenin subunits (Glu-3) are linked to genes (Gli-1) encoding omega and gamma gliadins. Because of this linkage between Gli-1 and Glu-3 loci identification the Glu-3 allele can be enhanced through analysis of Gli-1 encoded omega gliadins using SDS–PAGE (Galili and Feldman, 1984; Cornish and Lukow, 1996).

In Ethiopia, landraces still take the largest share of wheat production. The knowledge of the end-use quality of these landraces is limited. As Ethiopia is a centre of diversity for tetraploid wheat (Vavilov, 1929), there is great scope to find useful novel variability for various traits in this germplasm. Cataloguing of cultivated and wild germplasm has provided a number of novel alleles at different loci controlling protein subunit composition (Payne *et al*., 1984; Randhawa *et al*., 1995, 1997). A number of these alleles have been found to be associated with high gluten strength (Payne *et al*., 1984; Harjit-Singh *et al*., 2000). Such germplasm can be potential donor parents to improve the gluten strength and ultimately processing quality of durum wheat cultivars. Moreover, these novel alleles when transferred to hexaploid wheat could be helpful to improving the bread-making quality of bread wheat.

The objective of this study was to evaluate and characterize the Ethiopian tetraploid wheat germplasm for protein compositions particularly for gliadin, and HMW and LMW glutenin subunit composition.

Materials and methods

Plant materials

One hundred and twenty tetraploid wheat accessions (an accession represents collections of the original landraces from the same area at the same time), representing 110 landraces and 10 released varieties were used. The landraces were obtained from seeds that have been collected by the Institute of Biodiversity Conservation (IBC) of Ethiopia. These are collections from different major wheat-producing regions and different altitudes of Ethiopia from the Arsi, Bale, Gojam, Gonder, Shewa, Tigray and Wello regions, whereas the released varieties were obtained from Debre Zeit Agricultural Research Center (DZARC). Sport, Swedish hexaploid wheat, was used as reference in analysing both the HMW glutenin subunits and gliadins. The tetraploid wheat Claro De Balazote, Langdon and Mexicali, obtained from Dr Nieto-Taladriz (from the University of Polytechnic, Madrid, Spain) were used as standards for LMW glutenin subunits.

Analysis of HMW and LMW glutenin subunits, and D-zone omega gliadins

The HMW were extracted from individually ground grains and the proteins were separated according to the method of Payne *et al.* (1980) with some modifications according to Uhlen (1990) on 10% polyacrylamide gels in the presence of sodium dodecyl sulphate (SDS–PAGE). In order to identify whether the samples contain subunit 2* or not, a further SDS–PAGE analysis was performed using 8% gels.

The LMW subunits were extracted from ground half grains and the proteins were separated on 10% polyacrylamide gels in the presence of sodium dodecyl sulphate according to Singh *et al.* (1991).

After grinding half grains, D-zone omega gliadins were extracted and separated by SDS–PAGE according to Branlard *et al.* (1994) with modification according to Johansson (1996).

Staining

In order to classify the gluten composition, the gels were stained with Coomassie Brilliant Blue R-250 solution at least overnight according to Johansson *et al.* (1993) and de-stained in 8% (w/v) trichloroacetic acid (TCA) for a day.

Nomenclature

The nomenclatures or designations of Payne and Lawrence (1983), Nieto-Taladriz *et al.* (1997) and Khelifi *et al.* (1992) were used for the HMW and LMW glutenin subunits, and the D-zone omega gliadins, respectively. For the analyses of each of these three components, at least five grains from each of the accessions were used,

https://doi.org/10.1079/PGR2006110 Published online by Cambridge University Press

and then the pooled data were employed in the final analyses reported in the results.

Results

HMW glutenin subunits

The result on HMW and LMW glutenin subunits and D-zone omega gliadins is given in Table 1. Among the 120 accessions, 67 were homogenous for HMW glutenin subunit composition. In terms of species, the distribution of homogenous accessions was seven out of 11 in *Triticum aethiopicum*, six out of 10 in *Triticum dicoccon*, 49 out of 87 in *Triticum durum* and five out of 12 in *Triticum turgidum*. About 39% of the durum wheat contains Glu-A1x subunit. The HMW glutenin subunit 2****, not reported so far (Fig. 1), was common among the Ethiopian landraces. About 47% of the accessions possessed the new HMW glutenin subunit which was found only in the landraces. In only one accession of durum wheat was the 2* subunit observed. The most frequent subunits (in 58% of the accessions) encoded from chromosome 1B was 7 + 8 only, followed by 7 + 8/20 (28%) and subunit 20 was found in 25% of the accessions. Subunits such as 14 + 15 and 6 + 8 were observed in durum wheat (Table 1). A specific allelic variant of 7 + 8 was found (7 + 8 low), in which the band 7 shows a somewhat higher mobility compared to what is normal for 7.

LMW glutenin subunits

Seven out of 11, four out of 10, 52 out of 87 and six out of 12 accessions were homogenous in *T. aethiopicum*, *T. dicoccon*, *T. durum* and *T. turgidum*, respectively (Table 1). A total of 15 alleles encoding LMW glutenin subunits were found in the materials used for this study, and of these four, nine and two alleles corresponded to Glu-A3, Glu-B3 and Glu-B2 loci, respectively (Fig. 2). At the Glu-A3 locus each accession possessed one to three bands. The Glu-A3a allele that encodes band number 6 was present in 63% of the accessions homogenously. The allele Glu-A3b that encodes one subunit numbered 5 was present in 7.5% of the accessions. Ten and 7.5% of the accessions, respectively, were heterogenous containing Glu-A3a and Glu-A3e, and Glu-A3a and Glu-A3b. One accession contained only Glu-A3g encoding three bands. Three accessions contained Glu-A3a and Glu-A3g.

At the Glu-B3 locus, each accession possessed four or five bands of different mobility. In total, nine different alleles were observed. Of these, Glu-B3c, Glu-B3d and Glu-B3e alleles encode five bands each, whereas the others (i.e. Glu-B3a, Glu-B3b, Glu-B3f, Glu-B3g, Glu-B3h and Glu-B3i) encode four bands each. Glu-B3g was found in 39 and 23% of the accessions homogenously and in combination with other patterns, respectively. Only two allelic variants were detected at the Glu-B2 locus. The Glu-B2a allele encoding subunit 12 was found in 23% of the accessions. The null allele (Glu-B2b) was present in 56% of the accessions.

Omega gliadins

Amongst the 120 tetraploid wheat accessions analysed, nine D-zone omega gliadins were observed. In seven accessions there was not any band, and six of the accessions were homogenous for the maximum number of bands (six bands). The genetic combination of the 12 alleles coding for D-zone gliadins gave 35 different patterns. The most frequent patterns noted in 53% of the accessions were $d_1d_5d_6$, d_4, $d_1d_5d_6d_9$ and $d_1d_5d_6d_8d_9d_{10}$ (Table 1). The patterns of allelic presentation for the D-zone gliadins are given in Fig. 3.

Discussion

Knowledge about genetic diversity and the relationship of germplasm among breeding materials is useful in crop improvement strategies. As SDS–PAGE of glutenins and gliadins provides an easy tool for the allelic identification of storage proteins encoded at loci on group 1 chromosomes, the use of storage proteins can reveal differences within and among accessions at the molecular level providing a more direct, reliable and efficient tool for the conservation and management of germplasm. Our results indicated that the storage proteins are highly polymorphic, informative and useful to estimate variation within and among accessions, and among species of the Ethiopian tetraploid wheat. The number of bands per accession was found to be similar to that reported in durum wheat by previous workers (Gupta and Shepherd, 1988; Nieto-Taladriz *et al.*, 1997), although frequencies of different banding patterns were found to differ in the Ethiopian tetraploid wheat compared to those found in durum wheat in earlier investigations. For example, the g allele of the Glu-B3 locus in the present tetraploid wheat material was the most common allele unlike the results of Nieto-Taladriz *et al.* (1997) and Igrejas *et al.* (1999). Regarding the locus Glu-B2, the allele b was more frequent than the allele a in the present material, which was the reverse to what prevailed in the materials of Nieto-Taladriz *et al.* (1997) and Igrejas *et al.* (1999).

Table 1. Allelic composition at Glu-A1, Glu-B1, Glu-A3, Glu-B3, Glu-B2, Gli-A1 and Gli-B1 for 120 accessions in terms of species

		HMW glutenin subunits			LMW glutenin subunits				Omega gliadins		
Accession no./varieties	Species	Hm/Ht	Glu-A1	Glu-B1	Hm/Ht	Glu-A3	Glu-B3	Glu-B2	Hm/Ht	Gli-A1	Gli-B1
Arendeto[a]	*T. durum*/rv	Ht	2****/0	7 + 8	Ht	a	e/g	a/b	Ht	e/b	h/a
Asassa	*T. durum*/rv	Ht	0	6 + 8/7 + 8	Hm	e	g	a	Hm	e	e
Boohai	*T. durum*/rv	Ht	0	7 + 8/20	Ht	b	a	a/b	Ht	e	c/e
DZ 1640	*T. durum*/rv	Hm	0	20	Hm	a	a	a	Hm	e	e
Foka	*T. durum*/rv	Hm	0	20	Hm	g	a	a	Hm	e	a
Ginchi	*T. durum*/rv	Hm	0	20	Hm	a	a	a	Hm	e	g
Klinto	*T. durum*/rv	Ht	0	7 + 8	Hm	a	a	a	Hm	e	e
Quamy	*T. durum*/rv	Hm	0	20	Hm	e	g	a	Hm	e	c
Ude	*T. durum*/rv	Hm	0	7 + 8	Ht	a/e	a/g	a	Hm	e	f
Yerer	*T. durum*/rv	Hm	0	20	Ht	a/e	a/b	a/b	Ht	e	c/e
5071	*T. durum*	Hm	2****	20	Hm	a	g	b	Hm	e	a
5163	*T. durum*	Hm	0	7 + 8	Hm	a	f	b	Ht	e	j/c
5170	*T. durum*	Hm	0	7 + 8	Hm	b	a	a	Hm	e	j
5550	*T. durum*	Ht	0	7 + 8/20	Ht	a/b	c/g	b	Ht	e/b	j/a
5613	*T. durum*	Hm	0	20	Hm	a	g	b	Hm	e	a
5632	*T. durum*	Hm	0	7 + 8	Hm	a	h	a	Hm	e	a
5736	*T. durum*	Hm	2****	7 + 8	Ht	b	b/g	b	Hm	e	e
5917	*T. durum*	Ht	0	7 + 8/20	Ht	b	a/i	a/b	Ht	e/b	j/b/g
5926	*T. aethiopicum*	Ht	2****/0	7 + 8	Ht	a	d/f/g	b	Ht	e/b	j/a
6078	*T. aethiopicum*	Hm	0	7 + 8	Hm	e	e	a	Hm	d	c/a
6138	*T. diccocon*	Hm	2****	7 + 8	Hm	b	i	b	Hm	e	a
6222	*T. diccocon*	Ht	2****/0	7 + 8/20	Ht	a/b	a/e/i	b	Ht	b	j/a
6232	*T. durum*	Hm	2****	7 + 8	Ht	a	b/d	b	Ht	a/b	b
6856	*T. durum*	Hm	2****	7 + 8	Ht	a	b/g	a/b	Ht	e/a/b	b/a
6861	*T. durum*	Ht	0	7 + 8/20	Ht	a/g	b/g	b	Ht	a/d	a
6917	*T. durum*	Ht	2****/0	7 + 8/20	Ht	a/e	g/f	b	Hm	d	a
6927	*T. durum*	Ht	0	7 + 8/20	Hm	a	g	b	Ht	e	j/a
7027	*T. durum*	Hm	0	7 + 8	Ht	a/e	g	a/b	Ht	d	j/c/a
7119	*T. durum*	Hm	0	7 + 8	Ht	a/b	a/f	a/b	Hm	b	a
7197	*T. durum*	Ht	2****/0	20	Ht	a/b	a/f	a/b	Ht	e	j/c/a/e
7301	*T. durum*	Ht	2****/0	7 + 8/20	Ht	a	b/g	a/b	Hm	e	a
7369	*T. durum*	Ht	2****/0	7 + 8	Ht	a/g	b/g	b	Hm	e	a
7412	*T. durum*	Ht	2****/0	7 + 8	Hm	a	f	b	Hm	e	a
7508	*T. durum*	Ht	2****	7 + 8/20	Ht	a/e	e/g	a/b	Hm	e	a
7559	*T. diccocon*	Hm	2****	7 + 8	Hm	b	i	b	Hm	e	a
7563	*T. durum*	Hm	0	7 + 8	Ht	a/e	b/g	a/b	Ht	e/b	c/a
7692	*T. diccocon*	Hm	2****	20	Ht	a/e/	c/i	b	Ht	e/b	c/a
7927	*T. durum*	Ht	0	7 + 8/20	Hm	b	a	b	Hm	e	c
7935	*T. durum*	Hm	0	20	Hm	a	f	b	Hm	e	c
7943	*T. durum*	Ht	2****	7 + 8/20	Hm	a	a	b	Hm	e	a
7955	*T. durum*	Hm	0	7 + 8	Hm	a	a	b	Hm	e	a
8000	*T. durum*	Hm	0	20	Hm	a	g	b	Hm	e	a
8233	*T. turgidum*	Ht	0	7 + 8/20	Ht	a	a/g	a/b	Hm	e	a

https://doi.org/10.1079/PGR2006110 Published online by Cambridge University Press

Table 1. *Continued*

Accession no./varieties	Species	HMW glutenin subunits			LMW glutenin subunits				Omega gliadins		
		Hm/Ht	Glu-A1	Glu-B1	Hm/Ht	Glu-A3	Glu-B3	Glu-B2	Hm/Ht	Gli-A1	Gli-B1
8241	*T. turgidum*	Ht	2****/0	7 + 8	Ht	a/e	e/g	a/b	Ht	e	c/a
8291	*T. durum*	Hm	2****	7 + 8	Hm	a	g	b	Hm	e	a
8292	*T. durum*	Hm	2****	7 + 8	Hm	a	b	a	Hm	d	b
8356	*T. durum*	Ht	0	7 + 8/20	Hm	a	g	a	Ht	e	j/c/c (low)
203790	*T. durum*	Hm	0	20	Hm	a	a	a	Hm	e	j
203922	*T. durum*	Hm	0	20	Hm	a	c	b	Hm	e	j
203942	*T. durum*	Hm	0	20	Hm	a	g	a	Ht	a/b	a
203958	*T. durum*	Hm	0	20	Hm	a	b	b	Hm	d	b
204340	*T. aethiopicum*	Hm	0	7 + 8	Hm	a	i	b	Hm	e	c
204388	*T. turgidum*	Hm	0	7 + 8	Ht	a/b	f	a	Hm	e	c
208212	*T. durum*	Hm	2****	7 + 8	Hm	a	g	b	Ht	e	a/f
208218	*T. durum*	Ht	2*	6 + 8/7 + 8	Hm	a	g	b	Ht	e	a
208219	*T. durum*	Hm	2****	7 + 8	Hm	a	g	b	Hm	e	f
208233	*T. aethiopicum*	Ht	2****	7 + 8/20	Ht	a/b	a/g	b	Hm	d	a
208255	*T. durum*	Ht	2****/0	7 + 8	Hm	a	g	b	Ht	e/b	a
208267	*T. durum*	Ht	2****	7 + 8/20	Hm	a	b	a	Hm	e	c
208787	*T. durum*	Hm	0	20	Ht	a	a	a/b	Ht	e/c	j/a
210814	*T. turgidum*	Hm	0	7 + 8	Hm	b	g	a	Hm	e	c
212649	*T. durum*	Hm	0	14 + 15	Hm	a	g	b	Hm	e	c
212652	*T. turgidum*	Ht	0	7 + 8/20	Hm	a	g	a	Hm	d	j
214263	*T. diccocon*	Hm	0	20	Hm	b	i	b	Ht	e/d	j/a
214508	*T. durum*	Ht	2****/0	7 + 8/20	Ht	a/e	b/g	a/b	Hm	e	c
214512	*T. durum*	Hm	2****	7 + 8	Hm	a	g	b	Hm	e	a
214588	*T. durum*	Hm	2****	20	Hm	a	g	b	Hm	e	j
214591	*T. durum*	Ht	2****/0	7 + 8	Hm	e	f	b	Hm	b	c
219510	*T. diccocon*	Hm	0	7 + 8	Hm	a	g	b	Hm	e	a
222196	*T. turgidum*	Ht	2****/0	20	Ht	a	b/ g	a/b	Hm	d	j
222308	*T. aethiopicum*	Ht	2****/0	7 + 8	Ht	a	g/i	a/b	Ht	e	c/a
222333	*T. aethiopicum*	Hm	0	7 + 8	Ht	e	b/f	b	Ht	d	j/c
222339	*T. durum*	Hm	0	7 + 8	Hm	a	g	b	Hm	d	a
222344	*T. durum*	Hm	0	7 + 8	Ht	a	b/g	b	Hm	a	b
222350	*T. aethiopicum*	Hm	0	7 + 8	Hm	a	g	b	Hm	b	a
222386	*T. durum*	Hm	2****	7 + 8	Hm	a	g	b	Hm	b	a
222390	*T. turgidum*	Hm	2****	7 + 8	Hm	a	g	b	Hm	e	j
222477	*T. durum*	Hm	2****	20	Hm	a	g	b	Hm	e/d	j/b
222488	*T. durum*	Ht	2****	7 + 8/20	Hm	a	g	b	Ht	e	a/f
222489	*T. durum*	Ht	2****	7 + 8/20	Ht	a/b/e	f/g	a/b	Hm	e	e/a
222503	*T. durum*	Ht	2****/0	7 + 8	Ht	a/e	g	a/b	Hm	e	a
222533	*T. durum*	Ht	2****	7 + 8/20	Ht	a	b/f	b	Ht	b	e/a
222588	*T. durum*	Ht	0	7 + 8/20	Ht	a/e	g	a/b	Ht	e/b	a/a (low)
222627	*T. durum*	Hm	2****	20	Ht	a/e	b/g	a/b	Ht	e	c/a

https://doi.org/10.1079/PGR2006110 Published online by Cambridge University Press

Table 1. *Continued*

		HMW glutenin subunits			LMW glutenin subunits				Omega gliadins		
Accession no./varieties	Species	Hm/Ht	Glu-A1	Glu-B1	Hm/Ht	Glu-A3	Glu-B3	Glu-B2	Hm/Ht	Gli-A1	Gli-B1
222637	*T. durum*	Hm	2****	20	Hm	e	g	a	Ht	e/b	b/a
222681	*T. durum*	Hm	0	20	Hm	e	g	a	Hm	e	a
222702	*T. durum*	Hm	2****	7 + 8	Hm	a	g	b	Hm	e	a
222704	*T. durum*	Hm	2****	20	Ht	a	f/g	b	Hm	e	a
222713	*T. durum*	Ht	0	7 + 8/20	Ht	a	g	a/b	Hm	e	c
222796	*T. durum*	Ht	0	7 + 8/20	Ht	a/e	c/g/h	a/b	Hm	e	c
222799	*T. durum*	Hm	0	7 + 8	Hm	a	g	a	Hm	b	a
226119	*T. durum*	Ht	0	7 + 8/20	Hm	a	g	b	Hm	e	c
226166	*T. durum*	Hm	2****	20	Hm	a	g	b	Ht	d	c/a
226198	*T. durum*	Hm	0	20	Hm	a	g	a	Hm	d	a
226207	*T. turgidum*	Hm	0	7 + 8	Hm	a	g	b	Hm	e	c
226233	*T. durum*	Hm	0	7 + 8	Hm	a	g	a	Hm	e	j
226285	*T. turgidum*	Ht	0	6 + 8/7 + 8/20	Ht	a	a/b	b	Hm	b	b
226290	*T. durum*	Hm	0	7 + 8	Hm	a	d	b	Hm	e	c
226347	*T. dicccocon*	Hm	0	7 + 8	Ht	b	b/i	b	Hm	d	c
203766	*T. dicccocon*	Hm	2****	20	Hm	a	f	b	Hm	e	c
226365	*T. durum*	Ht	2****/0	7 + 8	Hm	a	g	b	Hm	b	a
226383	*T. durum*	Hm	0	20	Ht	b/e	b/f/g	b	Ht	e	c/f
226392	*T. durum*	Ht	2****	7 + 8/20	Ht	a/g	b/g	a/b	Ht	e	c/c (low)
226811	*T. aethiopicum*	Ht	2****/0	7 + 8	Hm	a	g	b	Ht	e/b	a
226825	*T. aethiopicum*	Hm	0	7 + 8	Hm	b	g	a	Hm	d	a
226836	*T. durum*	Ht	2****	7 + 8/20	Hm	a	g	b	Hm	e	a
226849	*T. durum*	Ht	0	7 + 8/20	Ht	b/e	a	a/b	Hm	e	j
226861	*T. aethiopicum*	Hm	0	7 + 8	Hm	a	g	b	Ht	e/b	f/a
226865	*T. dicccocon*	Ht	0	7 + 8/20/7 + 8	Ht	b	b/i	b	Hm	d	a
226914	*T. durum*	Ht	2****/0	7 + 8	Ht	a	g	a/b	Ht	e	c/a
226942	*T. diccocon*	Ht	2****	7 + 8/7 + 8 (low)	Ht	a/b	b/d	b	Ht	b/d	b/i
227058	*T. durum*	Hm	2****	20	Hm	e	b	b	Hm	e	c
231558	*T. aethiopicum*	Hm	0	7 + 8	Hm	a	g	b	Ht	e	i/a
231584	*T. durum*	Ht	0	7 + 8/20	Ht	a	b	b	Hm	b	c
231620	*T. durum*	Ht	2****/0	7 + 8/14 + 15/20	Ht	a/b	f/g	a/b	Ht	e/b	j/a/i
231623	*T. durum*	Ht	2****/0	7 + 8/14 + 15/20	Hm	a	g	b	Hm	e	a
238135	*T. turgidum*	Ht	2****/0	7 + 8/20	Ht	a	b/g	b	Ht	e	j/c
238136	*T. turgidum*	Ht	2****/0	7 + 8/20	Hm	a	b	b	Hm	e	c
238137	*T. turgidum*	Hm	2****	20	Hm	g	b	B	Hm	e	c
239708	*T. diccocon*	Ht	0	7 + 8/20	Ht	a/e	d/g/f	a/b	Ht	e	c/a

rv, released variety; Hm, homogenous; Ht, heterogenous. 'Low' indicates that the intensity of the band is less relative to the others.
[a] Arendeto is selected from landraces.

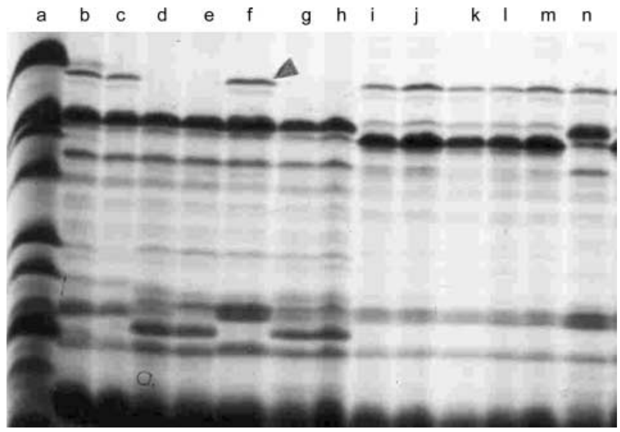

Fig. 1. SDS–PAGE separation of HMW glutenin subunits found in some tetraploid wheat accessions: (a) Sport, Swedish hexaploid wheat; (b, c) accession number 222390; (d–h) 208255; (i–m) 222477; (n) 222488. The arrowhead (▼) indicates the new allele designated as 2****.

between the different LMW models and allelic protein composition found for them (Nieto-Taladriz *et al.*, 1997), and it is also possible to establish equivalence models for our materials (Table 2). Branlard *et al.* (2003) have found that alleles Glu-A3a and Glu-A3d have a positive effect on the strength and extensibility of dough whereas allele Glu-B3g favours dough extensibility. It would be interesting to make a quality evaluation of the Ethiopian tetraploid wheat materials since the results of the previous and the present work showed that some of the accessions with the better allelic composition might have the highest sedimentation volume.

Many research programmes have shown that HMW and LMW glutenin subunits and polymerization of glutenins have a key role in the properties of gluten (Branlard *et al.*, 2001). The gliadins are not independent from gluten polymers; they are completely integrated into the complex net-

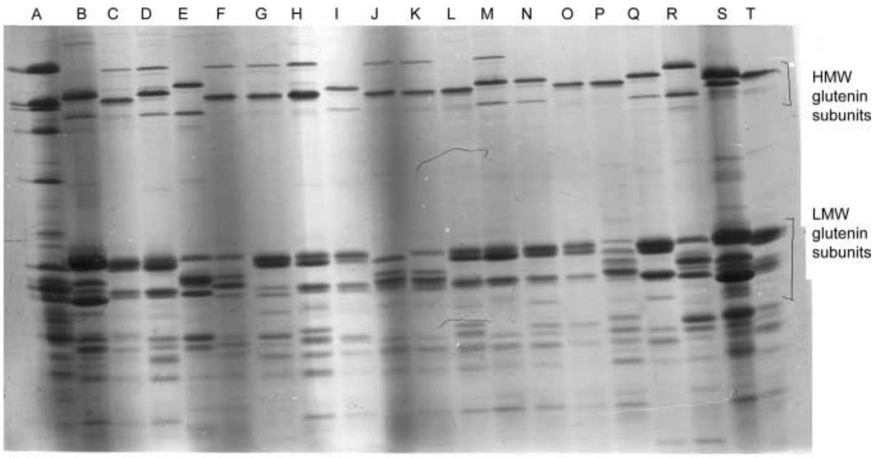

Fig. 2. Polymorphism of the glutenin subunits of the accessions of tetraploid wheat as revealed by SDS–PAGE: (A) Sport, Swedish hexaploid wheat; (B) Mexicali; (C) accession number 222477; (D) 222488; (E) Asassa; (F) 222489; (G) 222533; (H) 222704; (I) 222588; (J) 222627; (K) 222637; (L) 222681; (M) 222702; (N) 219510; (O) 222713; (P) 222796; (Q) 222799; (R) Langdon; (S) Claro de Balazote; (T) 226119.

Although the end-use quality analysis is based mainly on conventional technological tests, the use of SDS–PAGE may help to evaluate the quality of the tetraploid wheat. For instance, according to previous studies, durum wheat can be divided into groups depending on protein composition leading to differences in end-use quality of the wheat (Pogna *et al.*, 1990; Ruiz and Carrillo, 1995; Nieto-Taladriz *et al.*, 1997; Martinez *et al.*, 2004). According to these workers, those that possess LMW-2 and LMW-1 are associated with good and poor quality for the production of pasta, respectively. Even though the LMW-1 and -2 designations are inadequate since they represent a mixture of subunits controlled by different loci and alleles, protein compositions at the various LMW alleles describe better the relationships between proteins and quality in durum wheat. Equivalence models have been established

work forming the protein matrix more or less overlapping the starch granules (Branlard *et al.*, 2001, Kuktaite *et al.*, 2004). Gliadins are also important in providing viscosity

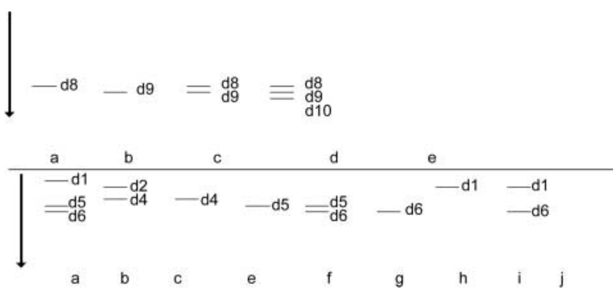

Fig. 3. Systematic presentation of the different allelic variants of D-zone omega gliadins of the 120 accessions as revealed by the SDS–PAGE technique.

https://doi.org/10.1079/PGR2006110 Published online by Cambridge University Press

Table 2. Equivalence of the allelic composition and the model of LMW glutenin subunits according to Payne *et al.* (1984) and Nieto-Taladriz *et al.* (1997)

LMW glutenin subunit model	Glu-A3	Glu-B3	Glu-B2
LMW-1	b	b	b
	b	i	b
	b	i	a
	b	b	a
LMW-2	a	a	a
	a	a	b
	a	g	b
	g	g	b
	b	a	a
	a	h	a
	e	e	a
	a	b	b
	g	a	a

and gluten extensibility which is a component of dough strength. Analysis of the individual and combined effects of LMW glutenin subunits, HMW glutenin subunits, and a fraction rich in albumins and globulins with some glutenins and omega gliadins has shown that LMW glutenin subunits, encoded by the Glu-B3 loci, and HMW subunits 7 + 8, encoded by the Glu-B1 locus on chromosome 1B are associated with high elastic recovery and gluten firmness and have a strong influence on gluten strength (Pogna *et al.*, 1990; Fares *et al.*, 1997).

Quamy (a released durum wheat cultivar in Ethiopia) possessed HMW glutenin subunit 20 and also the LMW-1 pattern which is associated with the poor quality of pasta products (Bechere *et al.*, 2002). Two other released cultivars, Boohai and Foka, also contained HMW glutenin subunit 20 but had the LMW-2 pattern that is associated with good quality of pasta products (Bechere *et al.*, 2002). The effect of the LMW and HMW glutenin subunits on gluten quality appears to be additive because accessions showing glutenin patterns LMW-2 in association with HMW glutenin subunits 6 + 8 and 7 + 8 were among those identified by the best gluten quality. This type of additive response was also reported by Boggini *et al.* (1997). As allelic identification of the storage proteins is very important for genotyping genetic resources and improving wheat quality, the further evaluation of the new banding pattern observed in the present tetraploid wheat material might help to associate it with the genetic resource and quality of the tetraploid wheat. In addition, the information presented may also be of interest to plant breeders for choosing parents to obtain recombinant lines with good end-use quality.

In Ethiopia, tetraploid wheat is used not only for the production of pasta but also for making bread, 'Injera' (also called leavened bread), traditional recipes, local alcohol, whole roasted or boiled seeds, porridge, etc. Consequently, our result showing the presence of differences in quality of the tetraploid can be due to the differences in the uses. To this end, *Triticum aethiopicum* is used for the preparation of local alcohol mainly and injera, whereas the *Triticum durum* wheat is used for the preparation of pasta products. *Triticum diccocon*, mainly used for the preparation of porridge and bread, revealed a combination of different storage proteins indicating a poor quality for pasta production (i.e. it possesses b, i, b or b, b, b composition of LMW glutenin subunits).

For all the tetraploid wheat species used for this study the B genome has been found more polymorphic than the A genome. This finding is in agreement with the result obtained by Alamerew *et al.* (2004) using microsatellites on Ethiopian tetraploid and hexaploid wheat. Ben Amer *et al.* (2001) also reported that the microsatellite loci of the B genome are more variable than those of the A genome.

In summary, the following major conclusions emerge from the results of our study. (i) Because the same HMW glutenin subunits can be associated with different patterns of LMW glutenin subunits, a substantial proportion of the variation in gluten properties can be explained in terms of gluten composition with LMW glutenin subunits making the largest contribution to determine the quality of the tetraploid wheat. (ii) Because the possibility of finding the small (rare) alleles in other material might be very low, the low frequency of some patterns may indicate the necessity of the protection and conservation of the accessions with such characters in order to decrease the loss of these alleles and maintain the genetic base of the wheat as a whole. (iii) New alleles involved in gluten properties are continually found in landraces, creating new opportunities and the need for additional analyses. (iv) In a relatively high proportion of the durum wheat material used for this study Glu-A1x was expressed which until now has been reported only in small proportions in durum wheat. (v) Correlation between the composition of gluten subunit patterns observed and pasta quality remain to be determined after quality evaluation of pasta produced from the different materials studied.

Acknowledgements

We are grateful to the Institute of Biodiversity Conservation and Debre Zeit Agricultural Research Center in Ethiopia for providing the plant materials used in this study and to Dr Nieto-Taladriz for providing us with the European tetraploid wheat which was used as a standard. We thank Marie-Luisa Prieto-Linde for assistance with the electrophoretic work and Kerstin Brismar for photographic work. We are grateful to Dr Kebebew Assefa for reviewing the manuscript. The work was supported by the Swedish International Development Agency (Sida/SAREC) for which we are very grateful.

References

Alamerew S, Chebotar S, Huang X, Röder M and Börner A (2004) Genetics diversity in Ethiopian hexaploid and tetraploid wheat germplasm assessed by microsatellite markers. *Genetic Resources and Crop Evolution* 51: 559–567.

Bechere E, Peña RJ and Mitiku D (2002) Gluten composition, quality characteristics, and agronomic attributes of durum wheat cultivars released in Ethiopia. *African Crop Science Journal* 10: 173–182.

Beitz JA, Shepherd KW and Wall JS (1975) Cereal single-kernel analysis of glutenin: use in wheat genetics and breeding. *Cereal Chemistry* 52: 513–532.

Ben Amer IM, Börner A and Röder M (2001) Detection of genetic diversity in Libyan wheat genotypes using microsatellite markers. *Genetic Resources and Crop Evolution* 48: 579–585.

Boggini G, Doust MA, Annicchiarico P and Pecetti L (1997) Yielding ability, yielding stability and quality of exotic durum wheat germplasm in Sicily. *Plant Breeding* 116: 541–545.

Branlard G, Dardevet M, Nieto-Taladriz MT and Khelifi D (1994) Allelic diversity of the omega gliadins as revealed by SDS-PAGE: their possible implication in quality variation. In: Lafiandra D, Masci S and D'Ovidio RD (Eds) *Gluten Proteins 1993*. Detmold, Germany: Association of Cereal Research, pp. 234–243.

Branlard G, Dardevet M, Saccomano R, Lagoutte F and Gourdon J (2001) Genetic diversity of wheat storage proteins and bread wheat quality. *Euphytica* 119: 59–67.

Branlard G, Dardevet M, Amiour N and Igrejas G (2003) Allelic diversity of HMW and LMW glutenin subunits and omega gliadins in French bread wheat (*Triticum aestivum* L.). *Genetic Resources and Crop Evolution* 50: 669–679.

Cornish GB and Lukow OM (1996) Relationship between low-molecular-weight (LMW) glutenin (Glu-3) alleles and D-zone omega gliadins (Gli-1). In: Wrigley CW (ed.) *Gluten '96: Proceedings of the 6th International Gluten Workshop*, pp. 408–413.

Damidaux R, Autran JC and Feillet P (1980) Intrinsic cooking quality evaluation in durum wheats through examination of gliadin electrophoregrams and measurements of gluten visco-elasticity. *Cereal Foods World* 25: 754–756.

Fares C, Novembre G, Di Fonzo N, Galterio G and Pogna NE (1997) Relationship between storage protein composition and gluten quality in breeding lines of durum wheat (*Triticum turgidum* ssp *durum*). *Agricoltura Mediterranea* 127: 363–368.

Galili G and Feldman M (1984) Mapping of glutenin and gliadin genes located on chromosome 1B of common wheat. *Molecular and General Genetics* 193: 293–298.

Galterio G, Grita L and Brunori A (1993) Pasta making quality in *Triticum durum*: new indices from the ratio among protein components separated by SDS-PAGE. *Plant Breeding* 110: 290–296.

Gupta RB and Shepherd KW (1988) Low-molecular-weight glutenin subunits in wheat: their variation, inheritance and association with bread making quality. In: Miller TE and Koebner RMD (eds) *Proceedings of the 7th International Wheat Genetics Symposium*. Bath: Bath Press, pp. 943–949.

Harjit-Singh, Dhaliwal HS and Yifru T (2000) Germplasm enhancement through wide hybridization and molecular breeding. In: *The Eleventh Regional Wheat Workshop for Eastern, Central and South Africa*. Addis Ababa, Ethiopia: EARO, pp. 25–34.

Igrejas G, Guedes-Pinto H, Carnide V and Branlard G (1999) The high and low molecular weight glutenin subunits and ω-gliadin composition of bread and durum wheats commonly grown in Portugal. *Plant Breeding* 118: 297–302.

Jackson EA, Holt LM and Payne PI (1983) Characterization of high-molecular-weight and low molecular-weight glutenin subunits of wheat endosperm by two dimensional electrophoresis and the chromosomal localization of their controlling genes. *Theoretical and Applied Genetics* 66: 29–37.

Johansson E (1996) Quality evaluation of D-zone omega gliadins in wheat. *Plant Breeding* 115: 57–62.

Johansson E, Henriksson P, Svensson G and Hennen WK (1993) Detection, chromosomal location and evaluation of the functional value of a novel high Mr glutenin subunit found in Swedish wheats. *Journal of Cereal Science* 17: 237–245.

Khelifi D, Branlard G and Bourgoin-Greneche M (1992) Diversity of some D zone omega gliadins of bread wheat as revealed by 2 step A-PAGE/SDS-PAGE technique. *Journal of Genetics and Breeding* 46: 351–358.

Kosmolak FG, Dexter JE, Matsuo RR, Leisle D and Marchylo BA (1980) A relationship between durum wheat quality and gliadin electrophoregram. *Canadian Journal of Plant Science* 60: 427–432.

Kovacs MIP, Howes NK, Leisle D and Zawistowski J (1995) Effect of two different low-molecular-weight subunits on durum wheat pasta-quality parameters. *Cereal Chemistry* 72: 85–87.

Kuktaite R, Larsson H and Johansson E (2004) Variation in protein composition of wheat flour and its relationship to dough behavior. *Journal of Cereal Science* 40: 31–39.

Liu CY and Rathjen AJ (1996) Association of high and low molecular weight glutenin subunits with dough strength in durum wheats [*Triticum turgidum* spp. *Turgidum* L. conv. *durum* (Desf.)] in south Australia. *Australian Journal of Experimental Agriculture* 36: 451–458.

Liu CY and Shepherd KW (1995) Inheritance of B subunits of glutenin and ω and γ-gliadins in tetraploid wheats. *Theoretical and Applied Genetics* 90: 1149–1157.

Martinez MC, Ruiz M and Carrillo JM (2004) New B low Mr glutenin subunit alleles at the Glu-A3, Glu-B2 and Glu B3 loci and their relationship with gluten strength in durum wheat. *Journal of Cereal Science* 40: 101–107.

Metakovsky EV, Novoselskaya AYu, Kopus MM, Sobko TA and Sozinov AA (1984) Blocks of gliadin components in winter wheat detected by one-dimensional polyacrylamide gel electrophoresis. *Theoretical and Applied Genetics* 67: 559–568.

Nieto-Taladriz MT, Ruiz M, Martinez MC, Vazquez JF and Carrillo JM (1997) Variation and classification of B low-molecular-weight glutenin subunit alleles in durum wheat. *Theoretical and Applied Genetics* 95: 1115–1160.

Payne PI and Corfield KG (1979) Subunit composition of wheat glutenin protein isolated by gel filtration in a dissociating medium. *Planta* 145: 83–88.

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

Payne PI, Law CN and Mudd EE (1980) Control by homoeologous group 1 chromosomes of the high-molecular-weight subunits of glutenin, a major protein of wheat endosperm. *Theoretical and Applied Genetics* 58: 113–120.

Payne PI, Jackson EA and Holt LM (1984) The association between γ-gliadin 45 and gluten strength in durum wheat varieties: a direct causal effect or the result of genetic linkage. *Journal of Cereal Science* 2: 73–81.

Pogna NE, Lafiandra D, Feillet P and Autran JC (1988) Evidence for a direct causal effect of low-molecular-weight glutenin subunits on gluten viscoelasticity in durum wheats. *Journal of Cereal Science* 7: 211–214.

Pogna NE, Autran JC, Mellini F, Lafiandra D and Feillet P (1990) Chromosome 1B-encoded gliadins and glutenin subunits in durum wheat: genetics and relationship to gluten strength. *Journal of Cereal Science* 11: 15–34.

Randhawa HS, Dhaliwal HS, Harjit-Singh and Haminda K (1995) Cataloguing of wheat germplasm for HMW glutenin subunit composition. In: Chopa VL, Sharma RP and Swaminatha MS (eds) *Second Asia-Pacific Conference on Agricultural Biotechnology*. New Delhi: Oxford and IBH Publishing Company, pp. 13–26.

Randhawa HS, Dhaliwal HS and Harjit-Singh (1997) Diversity for HMW glutenin subunit composition and the origin of polyploid wheat. *Cereal Research Communications* 25: 77–84.

Ruiz M and Carrillo JM (1995) Relationships between different prolamin proteins and some quality parameters in durum wheat. *Plant Breeding* 114: 40–44.

Shewry PR, Tatham AS, Forde J, Kries M and BJ (1986) The classification and nomenclature of wheat gluten proteins: a reassessment. *Journal of Cereal Science* 4: 97–106.

Singh NK, Shepherd KW and Cornish GB (1991) A simplified SDS-PAGE procedure for separating LMW subunits of glutenin. *Journal of Cereal Science* 14: 203–208.

Uhlen AK (1990) The composition of high molecular weight glutenin subunits in Norwegian wheats, and their relation to bread-making quality. *Norwegian Journal of Agricultural Science* 4: 1–7.

Vavilov NI (1929) Wheat of Ethiopia. *Bulletin of Applied Botany, Genetics and Plant Breeding* 20: 224–356.

Woychik JH, Boundy JA and Dimler RJ (1961) Starch gel electrophoresis of wheat gluten proteins with concentrated urea. *Archives of Biochemistry and Biophysics* 94: 477–482.