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Variation in High Molecular Weight Glutenin Subunits and Seed Chemical Contents in Ethiopian Bread Wheat (*Triticum aestivum* L.) Varieties

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ABSTRACT

High molecular weight glutenin subunits largely determine bread making quality of wheat. A study was done from April to May, 2015, in the Division of Plant Breeding, Department of Crop Sciences, University of Natural Resources and Life Sciences, Vienna, Austria, using seventeen bread wheat varieties of Ethiopia in order to determine high molecular weight glutenin subunits and seed chemical contents. Ash content ranged from 2.24 (variety Warrakatta/Pastor) to 3.44% (variety Mekelle-3), fat 2.73 (variety Alidoro) to 3.57% (variety Hulluka), starch 63.69 (variety Tay) to 70.98% (variety Alidoro), protein 14.00 (variety Mekelle-3) to 16.75% (variety Tay), gluten 32.96 (variety Mekelle-3) to 38.66% (variety Tay), and 1000-seed weight 24.30 (variety Jefferson) to 35.77(g) (variety Hidase). SDS-PAGE analysis showed that loci *Glu-A1*, *Glu-B1* and *Glu-D1* consisted of three, five and two subunits, respectively. Varieties ETBW5879, Worrakatta/Pastor, Hulluka, Mekelle-3, Kakaba, Danda'a, Alidoro, Pavon-76 and Jefferson consisted of subunits 1 or 2*, 7+8 or 17+18, and 5+10 at *Glu-A1*, *Glu-B1* and *Glu-D1* loci, respectively, and *Glu-1* quality score of ten. Variety Tay consisted of subunits null, 6+8 and 2+12 at *Glu-A1*, *Glu-B1* and *Glu-D1*, respectively, and *Glu-1* quality score of four. The present study suggests that varieties combining 1 or 2*, 7+8 or 17+18, and 5+10 subunits at *Glu-A1*, *Glu-B1* and *Glu-D1*, respectively, would be used to improve bread making quality of wheat in the future breeding programs.

INTRODUCTION

Wheat protein (8 to 20% of the grain) mainly contains albumins, globulins, gliadins and glutenins. Glutenin protein is a polymer of long chain of polypeptide subunits linked by inter polypeptide disulfide bonds. The reduction of these inter-chain bonds allows the separation of the subunits into high molecular weight (HMW) (80-130 kDa) and low molecular weight (LMW) (10-70 kDa) glutenin subunits (Bietz and Wall, 1972). The former constitutes about 5 to 10% and that of the later about 20 to 30% of the total grain protein (Eagles et al., 2002). The HMW glutenin subunits are encoded by two types of genes (x:y) that are located at *Glu-A1*, *Glu-B1* and *Glu-D1* loci close to centromere on the long arm of the homologous chromosomes 1A, 1B and 1D, respectively (Payne et al., 1982). The x-gene encodes for high molecular weight x-type subunit, whereas y-gene encodes for LMW y-type subunit (Shewry et al., 1992).

Bread wheat is expected to contain six different HMW glutenin subunits but due to silencing of some of these genes(x,y), most bread wheat cultivars possess three to five subunits. Thus, *Glu-A1* locus encodes for none or one active subunit (y-type of this locus is not expressed) and loci *Glu-B1* and *Glu-D1* encode for one or two x-type subunits each (Payne and Lawrence, 1983). On the other hand, genes encoding for LMW glutenin subunits are located on the short arms of homologous group 1 chromosomes at *Glu-A3*, *Glu-B3* and *Glu-D3* loci, and are tightly linked to *Glu-1* loci (Liu et al., 2010). Moreover, the presence or absence of specific HMW glutenin subunits largely determines bread making quality of wheat. In general, subunits null, 6+8, and 2+12 at *Glu-A1*, *Glu-B1* and *Glu-D1*, respectively, are negatively related to break making quality (Payne et al., 1987; Weegels et al., 1996). The present study was done to determine high molecular weight glutenin

subunits and seed chemical contents for seventeen Ethiopian bread wheat varieties.

MATERIALS AND METHODS

This study was done in the Division of Plant Breeding, Department of Crop Sciences, University of Natural Resources and Life Sciences, Vienna, Austria, from April to May, 2015, using seed samples of seventeen wheat varieties grown at Halaba experimental station (7°18'45"N, 38°06'49"E and 1765 m above sea level) of Hawassa Agricultural Research Centre, Hawassa, Ethiopia, from August to November, 2014, during main cropping season. Seed samples were analyzed for ash, fiber and starch contents using near-infrared reflectance spectroscopy (NIRS) (Buker, Ettlingen, Germany), and protein and gluten contents using NIR spectrometer, NIRFlex N-500 (BÜCHI, Labortechnik AG, Flawil, Switzerland). Seven seeds were sampled for each variety and ground into fine powder for the determination of HMW glutenin subunits using sodium dodecyl sulfate polyacrylamide gel electrophoresis (SDS-PAGE) with 10% polyacrylamide gels as it was described by Payne et al. (1980). The numbering of glutenin subunits and calculation of *Glu-1* quality score were done according to Payne and Lawrence (1983) and Payne et al. (1987), respectively.

RESULTS

Ash content ranged from 2.24 (variety Warrakatta/Pastor) to 3.44% (variety Mekelle-3), fat 2.73 (variety Alidoro) to 3.57% (variety Hulluka), starch 63.69 (variety Tay) to 70.98% (variety Alidoro), protein 14.00 (variety Mekelle-3) to 16.75% (variety Tay), gluten 32.96 (variety Mekelle-3) to 38.66% (variety Tay), and 1000-seed weight 24.30 (variety Jefferson) to 35.77(g) (variety Hidase) (Table 1).

Table 1. Five seed chemical contents and 1000-seed weight of seventeen Ethiopian bread wheat varieties

Variety	Ash%	Fat%	Starch%	Protein%	Gluten%	1000-seed weight (g)
Alidoro	3.17	2.73	70.98	15.22	36.06	29.32
Danda'a	2.51	3.19	68.14	14.63	34.67	29.63
ETBW5879	2.64	3.48	70.95	15.09	35.21	29.53
Hidase	3.16	3.25	65.2	15.41	36.52	35.77
Hoggana	3.06	3.14	69.25	14.34	34.22	25.94
Hulluka	2.69	3.57	66.24	15.17	35.31	26.89
Inseno-1	2.76	3.43	68.11	15.45	35.98	32.60
Jefferson	3.24	3.19	67.33	16.21	38.33	24.30
Kakaba	2.68	3.19	66.48	14.43	34.36	28.48
Mekelle-3	3.44	3.13	65.66	14.00	32.96	30.48
Mekelle-4	2.70	3.29	68.59	14.53	34.28	30.93
Ogolcho	2.89	2.74	68.61	14.21	33.97	29.85
Pavon-76	2.59	3.49	68.76	15.20	36.39	27.17
Shorima	3.06	2.86	70.14	15.32	36.04	28.43
Sofumar	2.92	3.10	69.48	15.02	36.13	27.21
Tay	3.01	3.42	63.69	16.75	38.66	24.68
Warrakatta/Pastor	2.24	3.10	68.93	15.76	36.99	27.50
Mean±SE	2.87±0.07	3.19±0.06	68.03±0.49	15.10±0.17	35.65±0.37	28.75±0.67

For the seventeen bread wheat varieties, SDS-PAGE analysis gave HMW glutenin subunits of three, five and two at *Glu-A1*, *Glu-B1* and *Glu-D1* loci, respectively. Out of the seventeen varieties, nine of them combined 1 or 2*, 7+8 or 17+18, and 5+10 subunits at *Glu-A1*, *Glu-B1*

and *Glu-D1*, respectively, and had *Glu-1* quality scores of ten. On the other hand, variety Tay consisted of null, 6+8 and 2+12 subunits at *Glu-A1*, *Glu-B1* and *Glu-D1*, respectively, and had the lowest *Glu-1* quality scores of four (Table 2).

Table 2. High molecular weight glutenin subunits and *Glu-1* quality scores of seventeen Ethiopian bread wheat varieties

Variety	<i>Glu-A1</i> subunit	<i>Glu-B1</i> subunit	<i>Glu-D1</i> subunit	<i>Glu-1</i> quality score
Alidoro	1	7+8	5+10	10
Danda'a	1	17+18	5+10	10
ETBW5879	1	17+18	5+10	10
Hidase	2*	14+15	2+12	?
Hoggana	1	7+9	5+10	9
Hulluka	1	17+18	5+10	10
Inseno-1	2*	7+9	5+10	9
Jefferson	2*	7+8	5+10	10
Kakaba	1	17+18	5+10	10
Mekelle-3	1	17+18	5+10	10
Mekelle-4	2*	7+9	5+10	9
Ogolcho	2*	7+9	5+10	9
Pavon-76	2*	17+18	5+10	10
Shorima	2*	7+9	5+10	9
Sofumar	2*	7+9	5+10	9
Tay	0	6+8	2+12	4
Worrakatta/Pastor	1	17+18	5+10	10

Allele designation of subunits involved a = 1 and 2+12, b = 2* and 7+8, c = null, d = 6+8 and 5+10, and i = 17+18.

DISCUSSION

As to the present study, the presence of HMW glutenin subunits of null, 1 and 2* at *Glu-A1* has been reported in previous studies in bread wheat (Payne and Lawrence, 1983; Payne et al., 1987; Lukaw et al., 1989; Nakamura, 2000; Anwar et al., 2003; Chaparzadeh et al., 2008; Giraldo et al., 2010) despite that Giraldo et al., (2010) have also reported the presence of 2** glutenin subunit. In the present study, the number of alleles obtained at *Glu-B1* was similar to that reported in previous studies (Payne et al., 1987; Galova et al., 2002; Anwar et al., 2003; Chaparzadeh et al., 2008) except that seven (Lukow 1989; Giraldo et al., 2010) and 11 (Nakamura, 2000) alleles have been reported by other studies. Even though there were two alleles observed in the present study at *Glu-D1*, three (Galova et al., 2002; Giraldo et al., 2010), four (Payne et al., 1987; Lukow et al., 1989), five (Anwar et al., 2003) and six (Nakamura, 2000)

alleles have been reported at this locus. Varieties having subunits 1 or 2*, 7 + 8 or 17 + 18, and 5+10 at *Glu-A1*, *Glu-B1* and *Glu-D1*, respectively, are expected to have high sedimentation and mixing time values which give high stability, elasticity and mixing tolerance to the dough compared to other subunits on the same locus (Giraldo et al., 2010).

In the present study, some varieties having 1 or 2* subunits at *Glu-A1* had reduced *Glu-1* quality scores attributed to 7+9 subunit at *Glu-B1* locus. The highest value of *Glu-1* quality score of ten was achieved for nine of the seventeen varieties studied indicating that these varieties could be expected for their good bread making qualities (Payne et al., 1987; Galova et al., 2002). However, variety Tay could be judged as weak quality genotype because of its null, 6+8 and 2+12 subunits giving it lowest *Glu-1* quality score of four. On the other hand, high positive correlation between protein and gluten contents ($r=0.97$, $p<0.01$) observed in the present

study could be expected because gluten represents 75 to 85% of the grain protein content (Abdel-Aal et al., 1996; Belderok et al., 2000). The present study suggests that varieties combining 1 or 2*, 7+8 or 17+18, and 5+10 subunits at *Glu-A1*, *Glu-B1* and *Glu-D1*, respectively, would be used to improve bread making quality of wheat in the future breeding programs.

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